# A MICROCOMPUTER CONTROLLED BIOREACTOR : A COMPARATIVE STUDY OF CONTROL SCHEMES FOR THE CONTROL OF DISSOLVED OXYGEN IN MOLASSES FERMENTATION

A Thesis Submitted
In Partial Fulfilment of the Requirements
for the Degree of

**MASTER OF TECHNOLOGY**

by

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to the

DEPARTMENT OF CHEMICAL ENGINEERING

**INDIAN INSTITUTE OF TECHNOLOGY, KANPUR**

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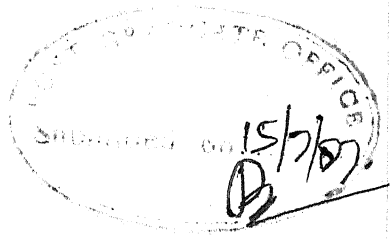

## CERTIFICATE

This is to certify that the work on 'A Microcomputer Controlled Bioreactor : A Comparative Study of Control Schemes for the Control of Dissolved Oxygen in Molasses Fermentation' has been carried out under our supervision and that this has not been submitted elsewhere for a degree.

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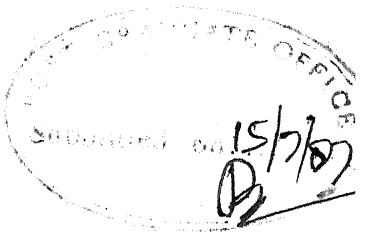

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-ASHWIN DESAI

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## NOMENCLATURE

| | | |
|---|---|---|
| $e$ | : | error (set point-measured variable), mg/l |
| $e_n$ | : | error at nth instant, mg/l |
| $K_c$ | : | proportional gain, $\frac{\text{number of steps}}{\text{mg/l}}$ |
| $K_p$ | : | process gain, $\frac{\text{mg/l}}{\text{number of steps}}$ |
| $t_d$ | : | dead time, s |
| $T$ | : | sampling rate, s |
| $v$ | : | controller output, steps |
| $v_o$ | : | steady state controller output, steps |
| $v_n$ | : | controller output at nth instant, steps |
| $\tau_D$ | : | derivative time constant, s |
| $\tau_I$ | : | integral time constant, s |
| $\tau_p$ | : | process time constant, s. |

## ABSTRACT

In the present work experimental studies have been done on the control of a bioreactor using a Personal Computer. Baker's yeast fermentation has been carried out under the controlled conditions of temperature and dissolved oxygen. An ON/OFF control scheme was used to control the temperature and a comparative study of Proportional (P), Proportional-Integral (PI) and Proportional-Integral-Derivative (PID) schemes was made for the control of the dissolved oxygen concentration. For the temperature, an ON/OFF control scheme was found to be satisfactory whereas for dissolved oxygen a Proportional-Integral-Derivative scheme was found to be the most suitable amongst the three control schemes which were considered. The present study was a part of the developmental work required to set up a low cost dedicated control system for the control of bio-reactors.

# CHAPTER 1

## INTRODUCTION

The development of the process of fermentation has historically been empirical, with the emphasis being on the systematic screening of medium ingredients, process conditions and new organisms. Systems commonly lacked feedback control of basic conditions such as temperature, pH, airflow, agitator speed, etc., few measurements were made other than the product. The modern philosophy of advancements through proper and thorough understanding demands entirely different equipment. Computers are increasingly becoming indispensable instruments to those engaged in fermentation research in maximizing the productivity and in widening the scope of observations as a result of shorter time lag between the occurrence of a physical event and its detection.

The use of computers for monitoring, operating and controlling fermentation processes has been rapidly increasing since the last fifteen years. Although the modern control theory was already well established by the 1960s, practical applications of the theory were not feasible until an adequate tool for its implementation, i.e. the computer, became available at a reasonable cost.

There are two main reasons for this widespread use of computers : affordability and necessity. Currently, the costs of computer logic and memory devices are plunging at a rapid rate and at the same time, the computational speed is also rapidly increasing, and the reliability is getting proportionately boosted. The advent of affordable microcomputers makes direct digital control economically more advantageous than analog control, even for a fermentation process of modest complexity.

Inspite of the widespread use of computers in fermentation processes there are still some serious hinderances to a fully computerized and automated fermentation process. One of them is the lack of adequate on-line sensors in monitoring both the physical and biological conditions inside a reactor, another is the lack of understanding of the microbial metabolic pathways and cellular control mechanisms, which is needed in order to model the process and formulate a meaningful process control algorithm so that the final objective of process optimization can be achieved.

Thus, the application of closed-loop control with a computer has been limited considerably because of the scarcity of reliable on-line sensors for monitoring key parameters in a fermentor. The succeeding work on modeling, control and optimization will proceed to the extent allowed by the development of the necessary sensors.

The present work involves a comparative study of three control schemes, viz. Proportional, Proportional-Integral, Proportional-Integral-Derivative for the control of dissolved oxygen in a bioreactor. The main objective was to develop a low cost, pc based control system which has been achieved. This study can be further used to develop a dedicated, microprocessor based control system which can be set up at a lower cost as compared to the pc-based system.

# CHAPTER 2

## LITERATURE REVIEW

The chemical process industry began utilizing direct digital computer control in the late 1950s. Since then the use of computer control in the chemical industries has been widespread. On the other hand, the idea of applying computers to control a fermentation process was proposed in the early 1960s and the fermentation industry has lagged behind the chemical industry in the application of the computers by approximately a decade or so.

In the 1940s there was the advent of digital computers, the forerunners of the computational machines that are in such widespread use today. Then, in 1945, there was simultaneously the development of submerged culture for use in antibiotics and especially penicillin production and the beginning applications of pH probes and gas analyzers for monitoring chemical processes. Although these events are isolated developments, they represent the foundation for the application of sensors, computers, and control theory to the development of modern fermentation technology.

By 1960 there was beginning to appear knowledge of how controlled addition of sugar feeds could be used to improve, in particular, antibiotic fermentation [1] . At this same time, the application of the theory for feed forward

control and direct digital control was introduced [2]. Then, in 1969 Yamashita et al. reported the first application of computer control to a fermentation process for the production of monosodium glutamate [3] . They reported that sequential control functions such as start-up, sterilization, and shutdowns were accomplished in pilot-scale and commercial batch glutamic acid fermentors by Ajinomoto Co. In this first application of a computer to a fermentation process temperature, pressure, pH, air flow rate, and foam were controlled and effluent gas composition, glutamic acid concentration, dissolved oxygen, and microbe density were monitored. Their approach to optimization was either by an early detection of change in the value of a principal parameter of the kinetic model of glutamic acid fermentation in each run or by comparing the performance of each fermentation with the standard run extracted from past experience. The latter approach is heuristic and requires no mathematical model.

Also, in 1969, Grayson [4] described the use of computer control in a fermentation plant with its application not only in the pilot plant but also in large-scale fermentors. He reported that direct digital control was conducted on 36 batch penicillin fermentation vessels with 114 control loops. The controlled variables were similar to those described by Yamashita et al.

In the 1970s there was a rapid proliferation of computer-coupled fermentation systems many of which were used in research and development programmes and some were used at the pilot plant scale.

In 1970, Henry [5] replaced the conventional analog controllers by a digital computer to accomplish complex control performance. The computer monitored the temperature of the culture broth, vessel pressure, pH of the broth, air flow rate and the addition of the antifoam agent.

All the work done during early 1970s was oriented towards data acquisition using the best available sensors, data analysis, development of computer programs for on-line control purpose, application of mathematical models and heuristic methods to resolve numerical and logical problems of cell growth, mutation and product formation. Finally, all this information helped in developing algorithms for on-line optimization prediction techniques and dynamic optimization.

There have been a few excellent review papers on the subject of computer control of fermentation processes at different stages of development. Nyiri [6] was among the first to write a review article. He examined the contributions prior to 1971. The publications he reviewed were mostly concerned with the off-line use of computers such as experimental data analysis, simulation, optimal trajectory

calculation, and parameter estimation for kinetic model. Only three papers cited in his review dealt with the use of computers for the purpose of process control. A substantial part of the article was devoted to various miscellaneous, but quite practical and essential considerations such as the development of models, the choice of computers and programming languages, methodology, and algorithms for data logging and analysis.

In 1977, Dobry and Jost [7] reviewed the developments published between 1972 and 1977 from an industrial point of view. They documented eleven existing fermentation systems interfaced to computers specifically for the purpose of monitoring and controlling a fermentor. Of the eleven systems being described, only two originated in industrial companies: the remaining were based in academic and institutional laboratories. Many articles discussed the application of digital computers to control fermentors on a real-time basis: however, most of them were of a highly speculative nature, and only very few dealt with the actual on line implementation of these ideas.

Finally, Dobry and Jost elaborated on the more practical industrial concerns such as the choice in the design of a computer-coupled fermentation system (types of control, data storage, computer, etc.) and the system requirements (documentation, data logging, computer languages for programming and the process control, etc.).

In the following year, Weigand [8] published a complementary review, emphasizing the developments in computer application to fermentation from an academic/research viewpoint. The system at Purdue University, U.S.A. was extensively documented. The paper reviewed the studies on modeling, control, and parameter estimation and optimization.

In 1979, Armiger and Humphrey evaluated the use of the indirect measurement concept [9]. The merit of using component balancing techniques for the on-line estimation of biomass concentration and growth rates and the possibilities of using these estimated values directly for control purposes were discussed. However, at the time, few studies were found to venture beyond the stages of on-line data acquisition and analysis.

In the same year, Zabriskie published a review which concentrated on the applications of computers principally for control purposes [10]. He also proposed an optimal multivariable control strategy for a fed-batch fermentor.

In 1982 Rolf and Lim reviewed the existing hardware software technology available for the implementation of uter control [11]. This paper's inclusion of on-line mation/identification and filtering aimed at feedback rol revealed the recent surge of interest toward on-line back control with the aid of sophisticated instrumen- on.

Hatch [12] provided a review of the latest advances in computer applications in the analysis of fermentor conditions and in the classical type of process control of fermentors. Bull [13] also surveyed the latest developments in computer-coupled instrumentation and the most recent studies in fermentation optimization.

Finally, Wang and Stephanopoulos presented a review of real-time digital computer applications to fermentation processes [14]. The state of instrumentation and the principles of operation were briefly examined with a special emphasis on the on-line capabilities. The advances in the application of modern computer control and optimization techniques to fermentation processes were discussed.

Thus, it can be seen that there are three main evolving stages in the use of computers — data acquisition, analysis, and control - and that each stage is progressively more difficult to solve.

## CONTROL OF DISSOLVED OXYGEN IN FERMENTATION PROCESSES

There is a wide variety of studies based on the measurement from a dissolved oxygen probe. The solubility of oxygen in fermentation broth [15] and the oxygen transfer coefficient under ideal conditions in a fermentor [16] have been studied. By comparing the desired set point to the dissolved oxygen concentration measured by an electrode, the control of dissolved oxygen level can be accomplished through

the manipulated changes in the agitation speed [17], aeration rate [18], or both [19].

As cells grow and multiply in a batch or fed-batch fermentor, the demand for oxygen increases, and dissolved oxygen can often become the growth limiting factor if it is not maintained properly. Automatic control of the dissolved oxygen level was attempted by Yano et al. [20], however, some control parameters had to be changed manually. Subsequent use of a microcomputer by Kobayashi et al. [21] and Nyiri et al. [22] successfully achieved the objective of keeping the dissolved oxygen at a constant level. For example, in Kobayashi et al.'s study of growing Candida Brassicae in a fed-batch mode, three variables (the agitation speed, the air flow rate, and the oxygen flow rate) were manipulated. A significant point in their approach was the use of a computer to determine the optimum choice of a set of operating points for the three manipulated variables such that the lowest running cost was realized. Although both aeration and agitation can be used to enhance the interfacial oxygen mass transfer, they exert different side effects on the fermentation broth, such as shear force and foam formation.

Two M.Tech. projects have been done in our lab on the on-line monitoring and control of bioreactors. P.K. Goel [23] worked on the 'Design and On-Line Computer Control of a Fermentor' in 1976. He used an ON/OFF control strategy to control the pH and the temperature of a Baker's Yeast fermentation process and utilised a mainframe computer (IBM 1800) for this purpose. With a view to reducing the cost of the control configuration, S. Kakkar [24] developed and tested a control system for a bioreactor using a microcomputer. He used an ON/OFF control strategy to control the pH and the temperature of the broth. The present study was done to control dissolved oxygen, which together with temperature and pH represents the three important parameters affecting the yield of a fermentation process.

Three, more sophisticated, control schemes viz. Proportional, Proportional-Integral, Proportional-Integral-Derivative were developed and tested for the control of dissolved oxygen. The present work was necessary to test and develop the control software which could then be used to set up a low cost, single board computer based dedicated control system.

# CHAPTER 3

# EXPERIMENTAL DETAILS

The complete experimental set-up consisted of:

1. The physical layout of the system,
2. The fermentation process,
3. The control scheme.

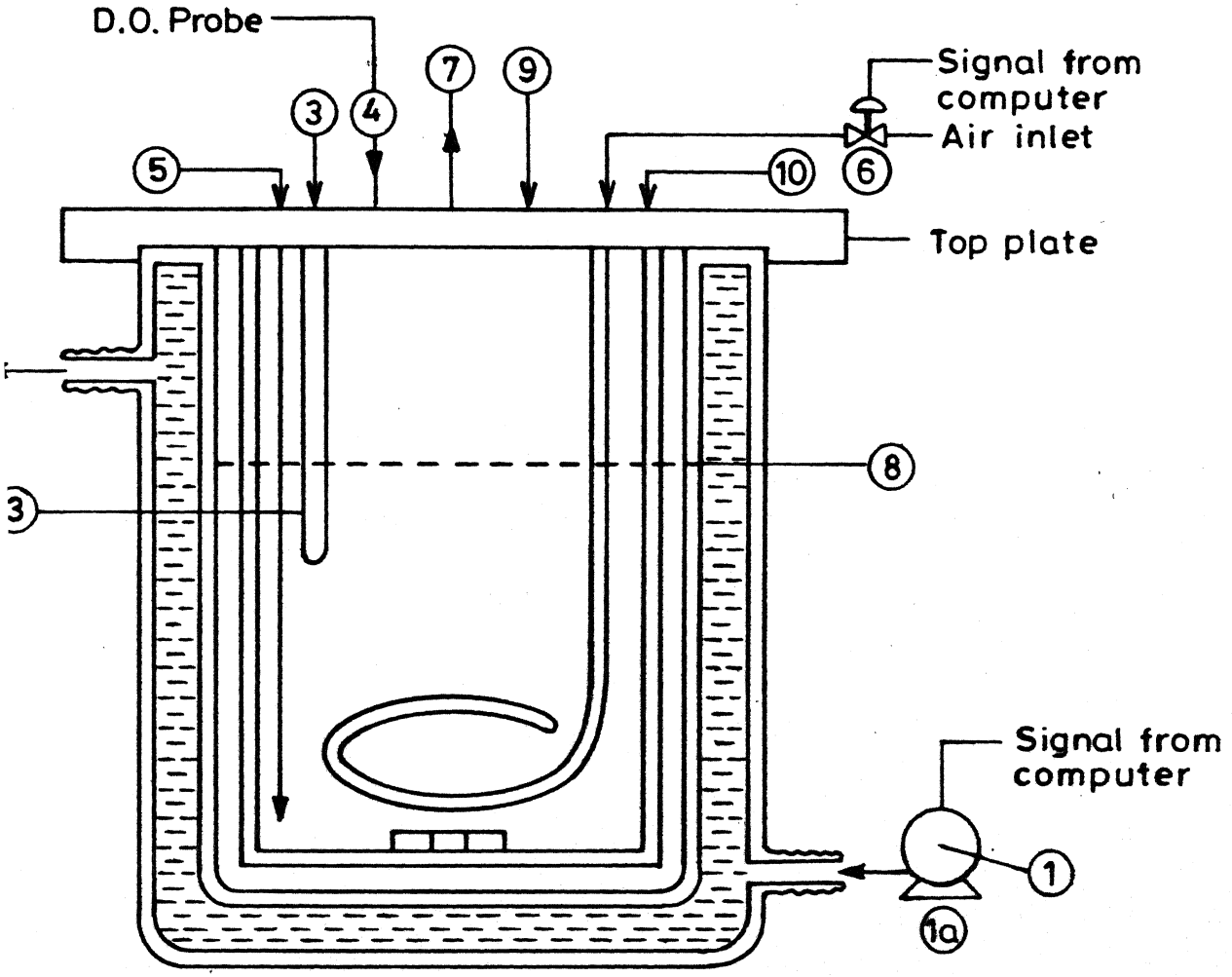
## 3.1 Physical Layout of the System

A 3-liter bioreactor was used for carrying out the fermentation. This bioreactor was fabricated by Sharma and Garg [25] for their senior project . It consisted of a double jacketted glass vessel wherein cooling water was circulated to maintain a constant temperature. Agitation was through a magnetic stirrer (Figure 1).

It was provided with a thermocouple (copper-constantan) for measuring the temperature and a polarographic-type Dissolved Oxygen Probe for measuring the dissolved oxygen of the broth. In addition, the system incorporated a compressed air supply consisting of a compressor, a filtering unit and a step motor driven needle valve for controlling the airflow to the bioreactor. It also included a constant temperature cooling bath and a cold water circulating pump to control the temperature of the bioreactor. Though the valves and actuators, and the microcomputer (Usha Eagle PC) were also part of the system, they have been discussed separately in

Key to Figure 1

| | |
|---|---|
| 1 | Coolant inlet |
| 1a | Cold water circulation pump |
| 2 | Coolant outlet |
| 3 | Thermocouple which measures temperature and sends signal to the computer |
| 4 | Dissolved oxygen probe which measures dissolved oxygen and sends signal to the computer |
| 5 | Steam inlet |
| 6 | Step motor controlled needle valve for manipulation of air flow rate |
| 7 | Air exhaust |
| 8 | Liquid level |
| 9 | Antifoam addition |
| 10 | Substrate feed. |




.1 Three-litre bioreactor with auxilary equipment.

Chapter 4 titled, 'Interfacing and Measurement'. A schematic representation of the bioreactor and its control scheme is shown in Figure 2.

## 3.2 Fermentation Process

Baker's yeast fermentation was carried out in the bioreactor. The volume of the liquid in the bioreactor was kept about 60% of the total volume because of the foam formation which would otherwise lead to overflow.

The complete process of the Baker's yeast fermentation involves three steps as given below.

### 3.2.1 Preparation of the Culture

The innoculum contains:

| Substance | % (W/V)* | Weight in gms. | Manufacturer |
|---|---|---|---|
| Yeast | 5% | 9 grams | The Indian Yeast Co. Ltd., Calcutta |
| Dextrose | 2% | 3.6 grams | I.D.P.L., Hyderabad |
| Peptone | 1% | 1.8 grams | I.D.P.L., Hyderabad |

(* Volume of culture = 180 ml).

The dextrose and peptone were dissolved together in distilled water and heated in a boiling water bath for 15 minutes in Pyrex bottles for sterilization. The

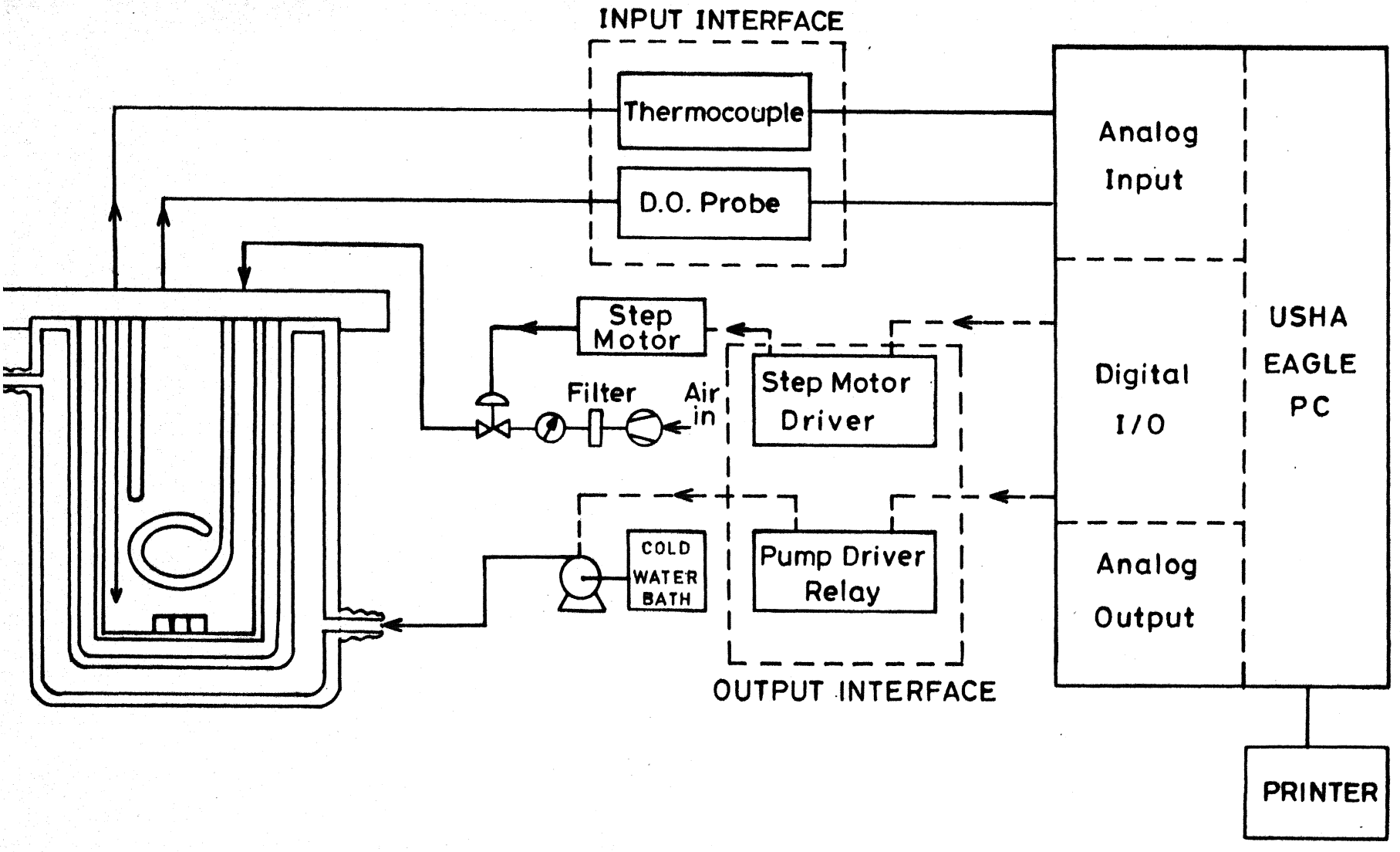



Fig. 2 Physical layout of system.

mixture was then cooled to room temperature by contact with a water stream. To this, yeast was added. Then distilled water was added to make the volume of the culture 1% (180 ml) of the total liquid volume. It was then plugged with cotton and kept in an incubator shaker at room temperature for 24 hours at 200 RPM.

### 3.2.2 Preparation of the Medium

The medium contains the following chemicals:

| Substance | % (W/V)* | Weight in gms. | Manufacturer |
|---|---|---|---|
| Substrate (Cane Molasses) | 5% | 90 grams | Obtained from N.S.I., Kanpur |
| Peptone | 1% | 18 grams | I.D.P.L., Hyedrabad |
| $MgSO_4$ | 0.5% | 09 grams | I.D.P.L., Hyderabad |

(* Volume of broth = 1.8 l ).

The peptone and $MgSO_4$ were first dissolved in distilled water in a 500 ml conical flask. The mouth of the flask was plugged with cotton. It was then heated in a boiling water bath for 10 minutes for sterilization. Similarly, the substrate was dissolved in distilled water and then sterilized separately. Then both were cooled to the room temperature. The medium was then transferred to the sterilized bioreactor.

### 3.2.3 Yeast Fermentation

The fermentor along with the stirrer and other auxiliaries were sterilized by rinsing with ethyl alcohol and leaving it overnight for 12 hours [26].

To start the fermentation, the medium was introduced into the bioreactor. To this, the culture containing the yeast was added and the total volume of the liquid in the bioreactor was adjusted to 60% of the total fermentor volume by adding distilled water. The agitation was then started. Initially the valve for letting in air was kept closed. This valve was then opened according to the demand for the dissolved oxygen.

Prior to the needle valve, air was passed through Purair filters (Manufactured by Shavo Norgren (India) Pvt. Ltd) to check contamination of microorganisms. There was also a master pressure regulator to control the pressure in the air line.

As the fermentation progresses, heat is released and thus the bioreactor temperature increases. To control the temperature at an optimum value of 28°C [27] the bioreactor was cooled by circulating cold water at a constant temperature through the jacket and thus the temperature was controlled to within $\pm$ 0.3°C. The control scheme for controlling the temperature including the circulating pump, and associated driver circuit was used by S. Kakkar [24] for his M.Tech. Project. Simultaneously, the dissolved oxygen of the

broth also started to decrease because of its consumption by the yeast cells. The dissolved oxygen was controlled by manipulating the airflow to the bioreactor by means of the stem position of the needle valve. To control the foam, ethyl acetate was added to the bioreactor.

The main reaction taking place was:

$$a\ C_xH_yO_z + C\ NH_3 + bO_2 \longrightarrow d\ C_eH_fN_gO_h + CO_2 + \cdots \quad (1)$$

(Substrate: Cane Molasses) (Yeast)

Fermentation of Baker's yeast being an aerobic reaction, the main byproducts were $CO_2$, water, ethanol and acids.

The reactions are as below:

$$C_6H_{12}O_6 \rightarrow 2\ C_2H_5OH + 2CO_2 \text{ (Anaerobic)} \quad (2)$$

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O \text{ (Aerobic)} \quad (3)$$

$$C_6H_{12}O_6 \rightarrow 2C_3H_6O_3 \rightarrow 2\ CH_3COCOOH + 2H_2 \quad (4)$$

(Hexose) (Triose) (Pyruvic Acid)

$$CH_3COCOOH \rightarrow CO_2 + CH_3CHO \xrightarrow{+H_2} C_2H_5OH \quad (5)$$

$$CH_3CHO + 2CO_2 + H_2O \longrightarrow C_2H_5OH + CH_3COOH \quad (6)$$

## 3.3 Control Scheme

The feedback control scheme consisted of the following:

(1) the process (Bioreactor)

(2) the measuring elements (Thermocouple, Dissolved Oxygen Probe)

(3) the on-line computer (Usha Eagle PC)

(4) the final control element (cold water circulating pump to control the temperature and a step motor controlled needle valve to control the dissolved oxygen).

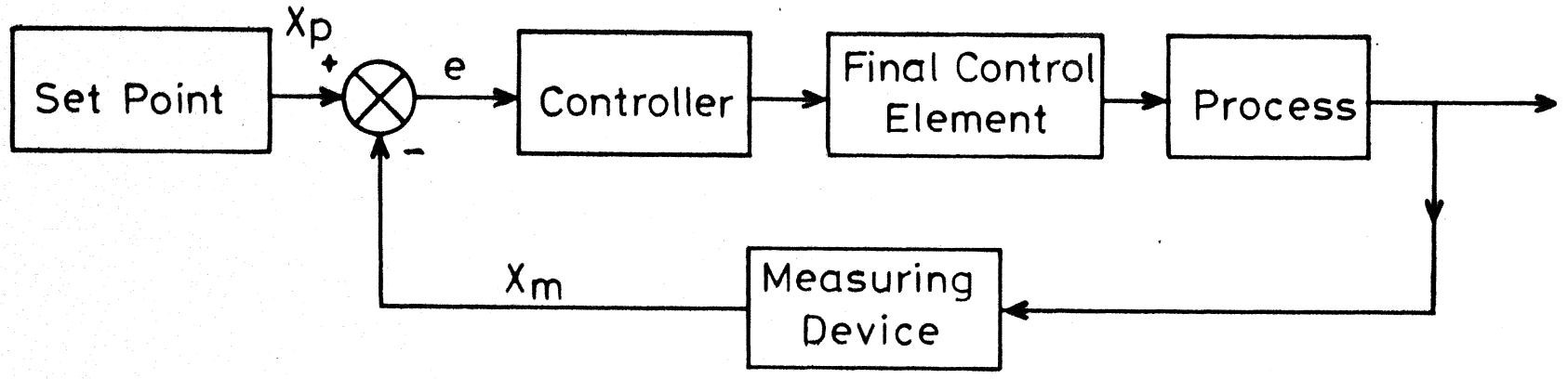
Figure 3 gives a block diagram for the control scheme in general. Two Single Input Single Output (SISO) control schemes were used to control the temperature and dissolved oxygen of the broth.

The control scheme for temperature was a closed loop ON/OFF type. The measured value was compared with the set point and the cold water circulation pump was switched 'ON' if the measured value was greater than the setpoint.

Three control schemes were used for controlling the dissolved oxygen content in the broth. They were - Proportional (P), Proportional + Integral (PI), Proportional + Integral + Derivative (PID).

Dissolved oxygen can be controlled by manipulating the airflow rate, the stirrer speed or a combination of the both. For the present work, it was controlled by manipulating the airflow rate while maintaining a constant stirrer speed.

The operation of an ideal PID controller is described by




Error $e = X_p - X_m$

**Fig. 3 Block diagram for a feedback control scheme.**

$$v = v_o + K_c \left( e + \frac{1}{\tau_I} \int_o^t e \, dt + \tau_D \frac{de}{dt} \right) \qquad (7)$$

where,

$v$ = controller output

$v_o$ = steady state controller output

$K_c$ = proportional gain

$e$ = error (set point-measured variable)

$\tau_I$ = integral time constant

$\tau_D$ = derivative time constant.

To obtain the digital equivalent to the PID controller, the derivative and the integral terms of the above equation are numerically approximated to give an expression for the output of the algorithm at the nth sampling instant. Thus,

$$v_n = v_o + K_c \left[ e_n + \frac{T}{\tau_I} \sum_{i=0}^{n} e_i + \frac{\tau_D}{T} ( e_n - e_{n-1} ) \right] \qquad (8)$$

where,

$v_n$ = controller output at nth instant

$v_o$ = steady state output of the control algorithm

$e_n$ = error at the nth sampling instant

$T$ = sampling rate

The velocity form of the PID algorithm is given below:

$$v_n = v_{n-1} + K_c \left[ (e_n - e_{n-1}) + \frac{T}{\tau_I} e_n + \frac{\tau_D}{T} (e_n - 2e_{n-1} + e_{n-2}) \right] \quad (9)$$

The velocity form was used for the control algorithm as it provided protection against reset windup, because it does not incorporate sums of error sequences.

The process gain ($K_p$), dead time ($t_d$), and process time constant ($\tau_p$) were obtained by the process reaction curve method as outlined in Appendix 4.

These 3 values, i.e. $K_p$, $t_d$ and $\tau_p$ were then used to estimate the controller parameters $K_c$, $\tau_I$ and $\tau_D$.

Cohen and Coon settings [28] were used to obtain the controller tuning. The equations used are given below:

(1) For proportional controllers

$$K_c = \frac{1}{K_p} \frac{\tau_p}{t_d} \left(1 + \frac{t_d}{3\tau_p}\right) \quad (10)$$

(2) For proportional-integral controllers

$$K_c = \frac{1}{K_p} \frac{\tau_p}{t_d} \left(0.9 + \frac{t_d}{12\tau_p}\right) \quad (11)$$

$$\tau_I = t_d \frac{30 + 3t_d/\tau_p}{9 + 20\, t_d/\tau_p} \quad (12)$$

(3) For Proportional-Integral-Derivative Controllers

$$K_c = \frac{1}{K_p} \frac{\tau_p}{t_d} \left(\frac{4}{3} + \frac{t_d}{4\tau_p}\right) \quad (13)$$

$$\tau_I = t_d \frac{32 + 6t_d/\tau_p}{13 + 8t_d/\tau_p} \quad (14)$$

$$\tau_D = t_d \frac{4}{11 + 2t_d/\tau_p} \quad (15)$$

The theoretical parameters obtained were used as a first guess and on-line tuning was then done to get the controller constants so that the fluctuations in the dissolved oxygen concentration in actual experiments are minimized. The controller output to the step motor was in terms of number of steps to be moved either in the reverse or in the forward direction and was given by:

(1) For Proportional Controller

$$v_n = v_{n-1} + K_c [(e_n - e_{n-1})] \quad (16)$$

(2) For Proportional-Integral Controller

$$v_n = v_{n-1} + K_c [(e_n - e_{n-1}) + \frac{T}{\tau_I} e_n] \quad (17)$$

(3) For Proportional-Integral-Derivative Controllers

$$v_n = v_{n-1} + K_c [(e_n - e_{n-1}) + \frac{T}{\tau_I} e_n + \frac{\tau_D}{T} (e_n - 2e_{n-1} + e_{n-2})] \quad (18)$$

The final controller parameters $K_c$, $\tau_I$, $\tau_D$ are:

(1) For proportional controllers: $K_c = 50$ (19)

(2) For proportional-integral controllers

$$K_c = 40 : \tau_I = 14 \quad (20)$$

(3) For proportional-integral-derivative controllers

$$K_c = 65 : \tau_I = 14.25 : \tau_D = 1.7836 \quad (21)$$

## 3.4 Constructional Details of the Bioreactor

### 3.4.1 Fermentation Chamber

The bioreactor was a double jacketted glass vessel which was 7.5 inch (19.05 cm) high and 5.5 inch (13.97 cm) in diameter; made by fusing the tops of two beakers of 3-litre and 5-litre capacity. The top was a 1/2 inch (1.27 cm) thick Perspex ring plate having a 5.5 inch (13.97 cm) inside diameter and 10.5 inch (26.67 cm) outside diameter. The inner side had a groove equal in diameter to that of the top of the beakers. It was fixed to the bioreactor with the help of epoxy resin (Araldite) to make it leakproof. The cold water was circulated through the jacket. The effective heat transfer area was 153.4 inch$^2$ (989.4 cm$^2$). The jacket was connected to a constant temperature bath.

### 3.4.2 Head Assembly Over the Top Perspex Ring

A 1/2 inch (1.27 cm) thick Perxpex Plate of 10.5 inch (26.67 cm) diameter was fixed which formed the top cover of the bioreactor.

### 3.4.3 Agitator

Agitation was done by a magnetic stirrer. For this a Remi-motor of 1/8 HP was used. The magnetic bar was a 1-inch (2.54 cm) piece.

### 3.4.4 Baffles

Four Perspex Baffles 6.25 inch (15.87 cm) in length, 1/2 inch (1.27 cm) width and 1/4 inch (0.64 cm) thick were provided all along the bioreactor height. At the bottom, they were attached to a reinforcing ring of Perspex by epoxy resin.

# CHAPTER 4

# INTERFACING AND MEASUREMENT

## 4.1 DT-2805 Card

For interfacing the Personal Computer to the bioreactor, a DT-2805 Low Level Analog and Digital I/O Card from Data Translation Inc., U.S.A. with its companion DT-707T screw terminal panel has been used. The DT system was used to monitor the temperature and dissolved oxygen and to actuate the pump for temperature control and to move the step motor in a forward/reverse direction to manipulate the stem position of the needle valve. It was also used to generate a 0.8V polarizing voltage for polarizing the dissolved oxygen probe. The specifications of the DT-2805 card are given in Appendix 2. It offers 8 Differential ADC channels with programmable gains of 1,10, 100 and 500, 2 DAC channels and 16 channels of Digital Input/Output.

The Digital Input Output section on DT-2805 has sixteen lines in two 8 bit blocks. These can be addressed separately or together. Either of the blocks can be configured as Input or Output. Thus, according to the requirements, it is possible to have 16 outputs or 16 inputs or 8 inputs and 8 outputs.

The Analog to Digital Converter is the soul of the system. It has a 12 bit ADC with 8 differential channels of input. As this is a low level card capable of accepting signals as low as 20 mV, differential inputs become necessary to eliminate the noise. The built-in software programmable amplifier permits gains 1,10, 100 and 500. The Digital to Analog Converter (DAC) has 12 bits of resolution on 2 channels. The DAC was used for the present system to generate an analog signal of 0.8V which is the polarizing voltage necessary for the dissolved oxygen probe.

All the outputs (Analog or Digital), hold the setting until it is changed or the board is reset. DMA operation is available throughout the analog section. There is also a facility to use an external trigger. The board has its own on-board clock and processor for fast operation. An external clock can also be used.

The DT card was installed in one of the long slots of the PC and draws about 1.2A at 5V from the mother-board. The DT-707T is the companion screw terminal panel for DT-2805 which was connected to it through a ribbon cable. All inputs and outputs are available on the DT-707T. The DT-707T also has a room temperature sensor on channel zero which can be disconnected if necessary. This is required if a thermocouple is being used. As the DT-707T has the temperature sensor on channel zero this channel is

not available unless the sensor is disconnected.

## 4.2 Measurement of the Variables to be Controlled

### 4.2.1 Dissolved Oxygen Measurement

Dissolved oxygen concentration in a fermentor is measured with an oxygen probe. There are two types of dissolved oxygen probes: (i) polarographic and (ii) galvanic depending on whether a polarizing voltage is required or not. These probes actually measure the activity, or the equivalent partial pressure, of the dissolved oxygen (oxygen tension) and not the concentration. The polarographic probe has the advantage of having a faster response and better sensitivity as compared to the galvanic probe and hence was used for the present work. The dissolved oxygen probe with its associated circuitry is given in Figure 4.

In a polarographic type of probe, a constant voltage is applied between the anode and the cathode and the current forced through the electrodes is measured. The oxidation-reduction reaction occurring at the electrodes is given below:

$$\text{Cathode} \quad \tfrac{1}{2} O_2 + H_2O + 2e \xrightarrow{Pt} 2OH$$

$$\text{Anode} \quad Ag + Cl \rightarrow AgCl + e$$

The dissolved oxygen probe (Yellow Springs Instrument Co., Ohio) consisted of a platinum cathode, silver anode,

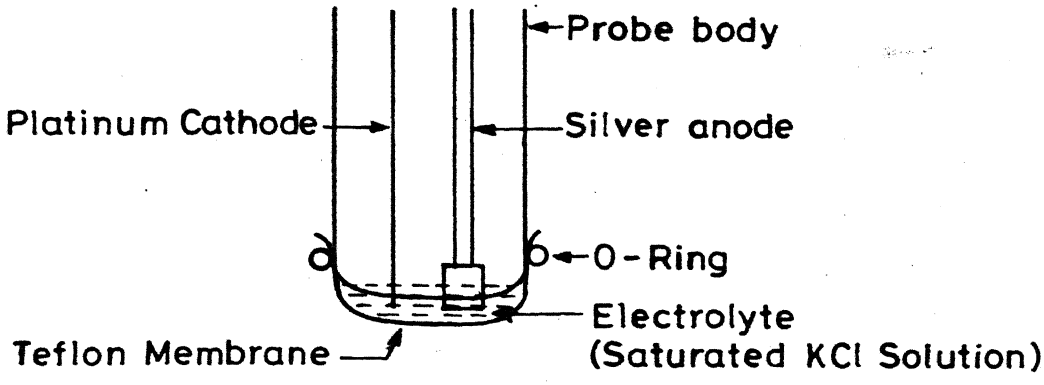



Fig. 4(a) Schematic of a dissolved oxygen probe.

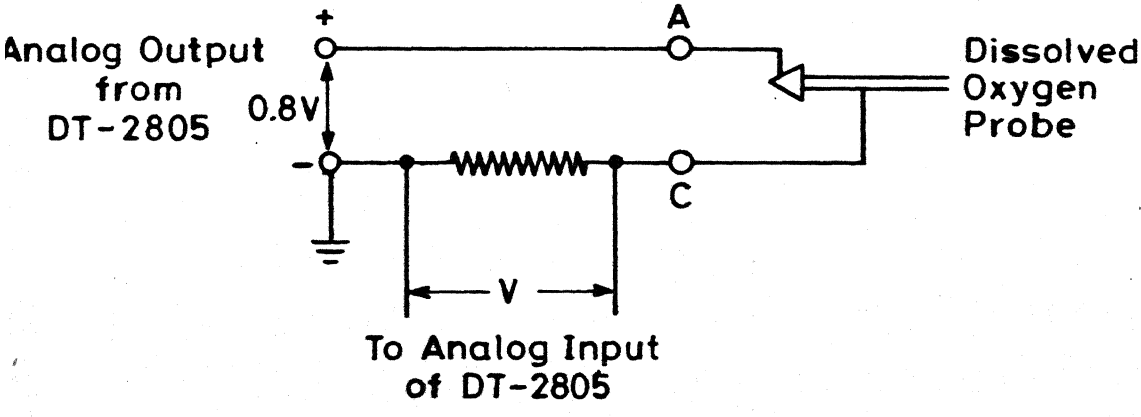



Fig. 4(b) Dissolved oxygen probe with associated circuit.

and KCl solution held captive around the electrodes by a Teflon membrane. The voltage versus dissolved oxygen calibration is given in Figure 16.

### 4.2.2 Temperature Measurement

The thermocouple was found to be the most convenient temperature device in the temperature range of our interest. The choice of a thermocouple is governed by the following aspects:

1. Higher emf/°C change in the temperature for higher accuracy in measurement. This improves both the sensitivity and signal to noise ratio.
2. Linearity in the range of operation.

Considering the above aspects, a copper-constanton (Type-T) thermocouple (Omega Inc.) . It gives a signal of around 40 microvolts/°C.

## 4.3 Output Interface

The temperature was controlled by manipulating the flow of cold water through the jacket of the bioreactor and the dissolved oxygen was controlled by manipulating the stem position of a step motor controlled needle valve.

A step motor is an electromagnetic incremental actuator which converts digital pulse inputs to analog output shaft motion. The step motor used to manipulate the

stem position of the needle valve had a rating of 2 kg-cm, 0.5A and 12V (manufactured by Srijan Drives, Pune). The output shaft of the motor rotates in equal increments to a train of input pulses. It moves 1.8°/step and thus has 200 steps/revolution. Step motors are constructed to facilitate solid state drive control, i.e. each winding requires current to be passed in one direction or no control at all. It has four windings which have to be energized in pairs. The four windings energize the stator so that the rotor advances one step at a time if the correct sequence or combination is used.

Thus, two different sets of pulse sequences were used for clockwise or anticlockwise motion.

Table 1 gives the sequence for clockwise motion:

| Step No. | I | II | III | IV |
|---|---|---|---|---|
| 1 | 1 | 1 | 0 | 0 |
| 2 | 0 | 1 | 1 | 0 |
| 3 | 0 | 0 | 1 | 1 |
| 4 | 1 | 0 | 0 | 1 |
| 1 | 1 | 1 | 0 | 0 |

Table 2 gives the sequence for anticlockwise motion:

| Step No. | I | II | III | IV |
|---|---|---|---|---|
| 4 | 1 | 0 | 0 | 1 |
| 3 | 0 | 0 | 1 | 1 |
| 2 | 0 | 1 | 1 | 0 |
| 1 | 1 | 1 | 0 | 0 |
| 4 | 1 | 0 | 0 | 1 |

Thus by providing the appropriate sequence of energizing the windings it is possible to have the output shaft motion in a clockwise or in an anticlockwise direction.

The TTL output from the DT-2805 cannot directly drive the motor. Thus, a driver is required. For safety reasons opto-coupling is desirable. For driving the step motor a driver circuit has been developed and is shown in Figure 5.

Electrical interferences was observed between the Dissolved Oxygen Probe and the other two sensors, viz. the thermocouple and the pH probe. As soon as the Dissolved Oxygen Probe was inserted into the broth, both the temperature and the pH signals went haywire. This was due to the electrical interconnection of the probes through the broth. The thermocouple was electrically isolated by placing it in a thin, closed glass tube. This slowed down its response

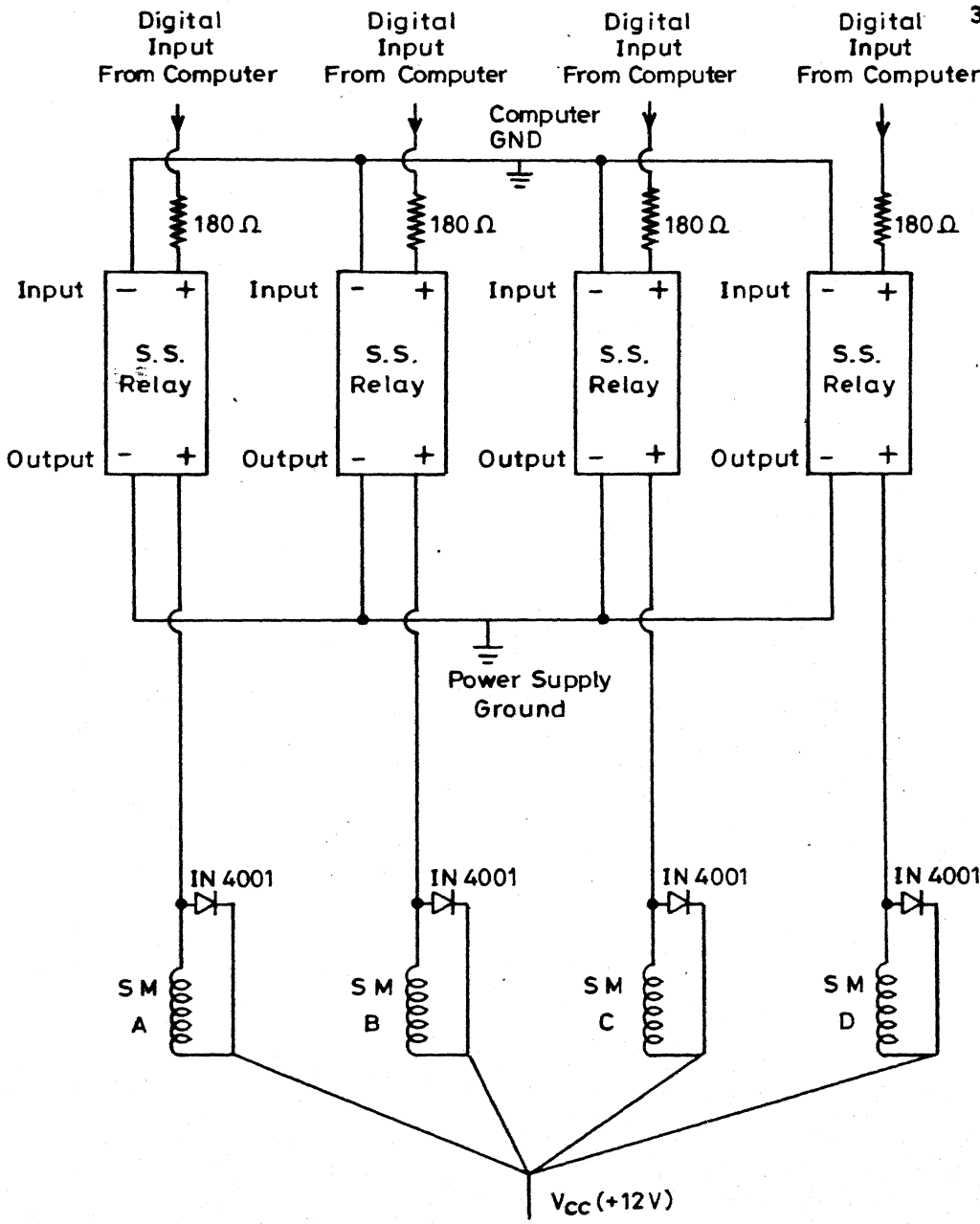



Fig. 5 Driving circuit for step motor.

somewhat but solved the problem. No such isolation was possible for the pH probe because the liquid must touch its glass bulb for measurement. Attempts to get round the problem by grounding either end of the Dissolved Oxygen Probe assembly proved fruitless. Thus it was not possible to run the Dissolved Oxygen Probe and the pH probe simultaneously. This shall be possible only if the Dissolved Oxygen Probe is isolated electrically.

This can be achieved through the use of an optically isolated interface in place of DT-707T. Alternatively the output of the dissolved oxygen probe can be used to drive a voltage to frequency converter which can be transformer coupled to a frequency to voltage converter which in turn is connected to the DT-707T. A galvanic probe, if used, would also eliminate the problem of interaction.

# CHAPTER 5

## SOFTWARE DEVELOPMENT

While it is quite possible to run the DT-2805 card through the BASIC INPUT and OUTPUT commands, it involves writing lengthy and tedious programs and hence it is not very convenient. A much more convenient approach is to use the software handler called PCTHERM from Data Translation. It can be addressed in BASIC and comes as two files: PCTHERM.LIB and PCTHERM.BAS. PCTHERM.LIB consists of routines in a library form for use with the BASIC compiler. It consists of a BASIC "save module", which contains a number of machine language routines designed to be called from IBM PC's compiled BASIC. It also consists of a set of stand alone data acquisition utilities. It has an error processing system which checks for the argument errors and generates an error report if any is detected. It greatly simplifies the programming required to use the DT card for a variety of applications. PCHTHERM.BAS is for use with the BASIC Interpreter and has been used for the present system. It is a collection of machine language routines that is designed to be loaded into memory by BASIC's BLOAD command. The BLOAD moves all the instructions into a section of memory that is otherwise not used. The starting address of this routine is set through the DEF SEG command.

PCTHERM requires 10 Kbytes of memory when it is used with a BASIC interpreter, it is usually loaded in the top 10 Kbytes of memory. In any system having 128 Kbytes or more of memory, PCTHERM is placed in the 10 Kbytes between 118 and 128 Kbytes. This is done by using a DEF SEG statement which is calculated as under:

128K - 10K = 118K

$118K = 118*2^{10} = 118*1024 = 120832$

120832/16 = 7552.

Thus, the commands to load PCTHERM at an offset of zero from 7552 are

```
DEF SEG = 7552
BLOAD "PCTHERM", Ø
```

The various routines used in the development of the software are given below. The input and output parameters are identified by capital and lower case letters respectively.

1. Reset.DT [XRD]

XRD (id %)

Entry point = 111.

This routine stops whatever the DT-2805 board is doing, perform a board reset, and reads back the identification code from the board id% : An integer identification code supplied by the DT-2805 board which indicates the board model number and microcode revision level. The integer consists

of a 16-bit field.

2. Measure.Thermocouple [XMT]

XMT (Type %, CHANNEL %, degrees.C!)

Entry point = 141.

This routine calculates the temperature Degrees.C! of a thermocouple of Type % connected on input channel channel %.

3. Measure.Volts [XMV]

XMV (CHANNEL %, Volts !)

Entry point = 138.

This routine performs a single A/D conversion on input channel %. It sets the input programmable gain of A/D board automatically to achieve maximum resolution without overloading the converter's input circuitry. The result of the conversion is an Analog Data Value which 'measure volts' converts to a single precision voltage value and returns as the variable volts!

4. Measure Cold Junction [XMCJ]

XMCJ (Degrees.C!)

Entry Point = 144.

This routine returns the temperature (in degrees celsius) of the DT-707T screw terminal panel. This temperature

can then be used to compensate for the error introduced by the cold-junction thermocouple formed where the thermocouple leads connect to the screw terminal panel.

5. Enable For Output [XEFO]

XEFO (Port. select %)

Entry Point = 060.

This routine enables either or both of the digital I/O ports for output. This routine has to be called before an output value can be set on the indicated port. Once a port is enabled for output, it remains so until explictly changed by the Enable for Input routine.

6. Output Digital Value [XODV]

XODV (PORT.SELECT %, MASK %, DIGITAL VALUE %)

Entry Point = 066.

This routine transfers the value of the bits in Digital Value to the digital output lines specified by Mask%.

7. Generate Volts [XGV]

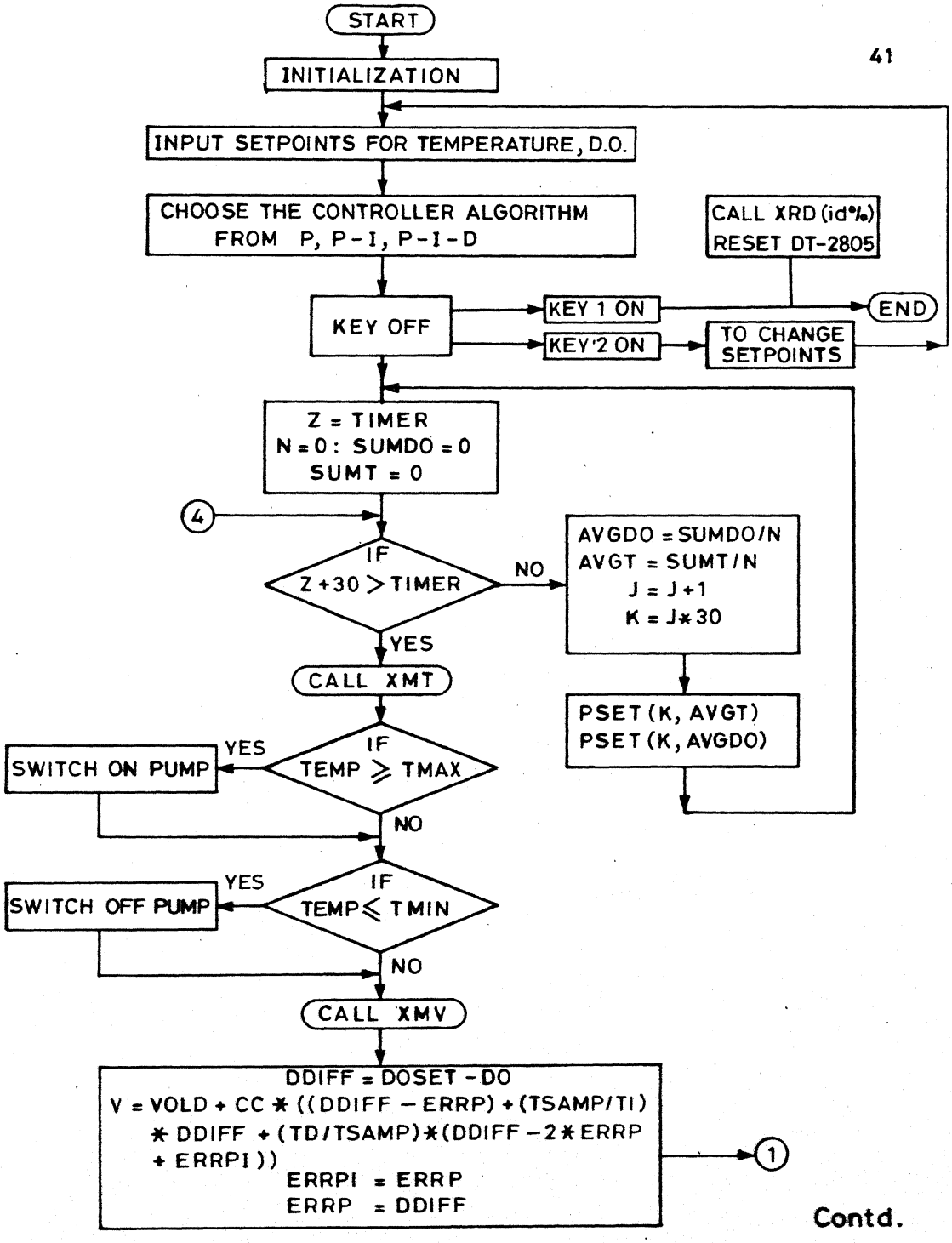
XGV (DAC.SELECT%,Volts !)

Entry Point = 165.

This routine generates the voltage indicated by volts! on the DAC (or DAC's) indicated by DAC.SELECT %.

Temperature and dissolved oxygen values are recorded on a floppy disc and aprinter. Also, on-line plotting is done with the help of BASIC commands. BASIC commands have been used for on-line plotting of temperature and dissolved oxygen with time, viz. VIEW and WINDOW. The VIEW statement defines a rectangular subset of the screen onto which the WINDOW contents are mapped. The WINDOW statement defines the world coordinate space that will be mapped into the physical coordinate space, as defined by the VIEW statement.

The listing of the program is given in Appendix 5. Also Figure 6 represents a flow diagram for the above control program. The data logging and results of temperature and dissolved oxygen are given in Table 3.


START
INITIALIZATION
INPUT SETPOINTS FOR TEMPERATURE, D.O.
CHOOSE THE CONTROLLER ALGORITHM FROM P, P-I, P-I-D
CALL XRD (id%)
RESET DT-2805
KEY OFF
KEY 1 ON
KEY 2 ON
END
TO CHANGE SETPOINTS
Z = TIMER
N = 0 : SUMDO = 0
SUMT = 0
4
IF Z+30 > TIMER
NO
YES
AVGDO = SUMDO/N
AVGT = SUMT/N
J = J+1
K = J*30
PSET (K, AVGT)
PSET (K, AVGDO)
CALL XMT
IF TEMP ≥ TMAX
YES
SWITCH ON PUMP
NO
IF TEMP ≤ TMIN
YES
SWITCH OFF PUMP
NO
CALL XMV
DDIFF = DOSET - DO
V = VOLD + CC * ((DDIFF - ERRP) + (TSAMP/TI) * DDIFF + (TD/TSAMP)*(DDIFF - 2*ERRP + ERRPI))
ERRPI = ERRP
ERRP = DDIFF
1

Contd.

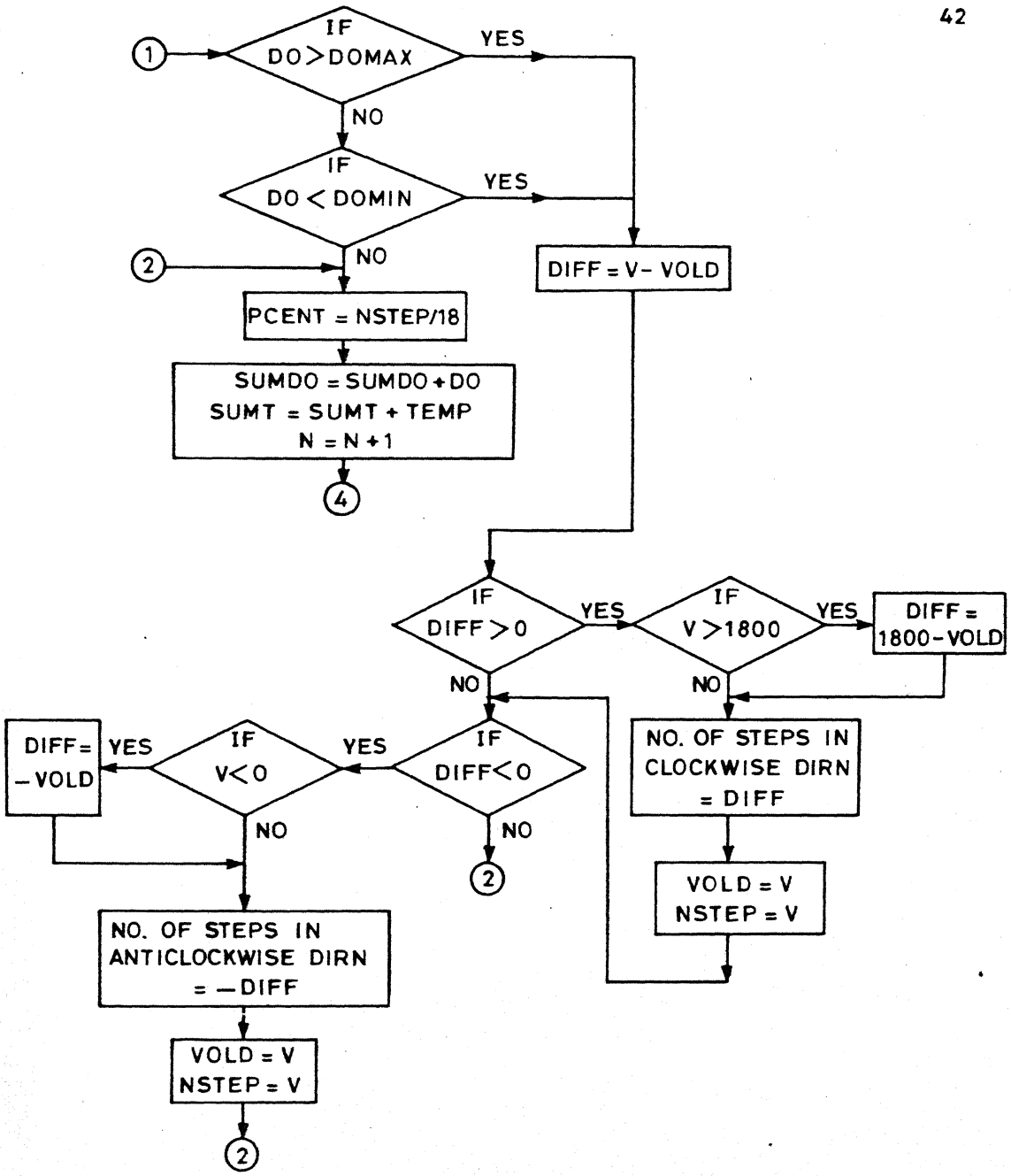



Fig. 6 Flow diagram for temperature and D.O. control.

prop :5th june
setpoints TEMPERATURE(C) 28 DEADBAND .6
DO 2.Ø DEADBAND .1

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| ØØ:22:Ø1 | TEMP | 28.3 ON | DO | 4.52 | NSTEP | Ø |
| ØØ:23:11 | TEMP | 27.6OFF | DO | 4.54 | NSTEP | Ø |
| ØØ:45:Ø5 | TEMP | 28.4 ON | DO | 2.48 | NSTEP | Ø |
| ØØ:47:14 | TEMP | 27.4OFF | DO | 2.49 | NSTEP | Ø |
| ØØ:59:32 | TEMP | 27.4OFF | DO | 1.94 | NSTEP | 3 |
| Ø1:ØØ:21 | TEMP | 27.4OFF | DO | 1.95 | NSTEP | 4 |
| Ø1:Ø1:4Ø | TEMP | 27.4OFF | DO | 1.95 | NSTEP | 4 |
| Ø1:Ø1:49 | TEMP | 27.4OFF | DO | 1.95 | NSTEP | 6 |
| Ø1:Ø2:Ø9 | TEMP | 27.9OFF | DO | 1.94 | NSTEP | 7 |
| Ø1:Ø2:14 | TEMP | 27.9OFF | DO | 1.92 | NSTEP | 8 |
| Ø1:Ø2:18 | TEMP | 27.7OFF | DO | 1.92 | NSTEP | 8 |
| Ø1:Ø2:22 | TEMP | 27.7OFF | DO | 1.94 | NSTEP | 6 |
| Ø1:Ø2:38 | TEMP | 27.9OFF | DO | 1.93 | NSTEP | 8 |
| Ø1:Ø2:42 | TEMP | 27.9OFF | DO | 1.93 | NSTEP | 8 |
| Ø1:Ø2:54 | TEMP | 27.6OFF | DO | 1.94 | NSTEP | 1Ø |
| Ø1:Ø2:59 | TEMP | 27.9OFF | DO | 1.93 | NSTEP | 11 |
| Ø1:Ø3:Ø3 | TEMP | 27.9OFF | DO | 1.93 | NSTEP | 11 |
| Ø1:Ø3:16 | TEMP | 28.2OFF | DO | 1.94 | NSTEP | 12 |
| Ø1:Ø3:28 | TEMP | 27.9OFF | DO | 1.94 | NSTEP | 13 |
| Ø1:Ø3:32 | TEMP | 27.9OFF | DO | 1.92 | NSTEP | 14 |
| Ø1:Ø3:4Ø | TEMP | 27.9OFF | DO | 1.95 | NSTEP | 15 |
| Ø1:Ø4:21 | TEMP | 28.1OFF | DO | 1.94 | NSTEP | 16 |
| Ø1:Ø4:25 | TEMP | 28.1OFF | DO | 1.93 | NSTEP | 17 |
| Ø1:Ø4:3Ø | TEMP | 27.8OFF | DO | 1.93 | NSTEP | 17 |
| Ø1:Ø4:38 | TEMP | 27.6OFF | DO | 1.93 | NSTEP | 2Ø |
| Ø1:Ø4:46 | TEMP | 27.9OFF | DO | 1.94 | NSTEP | 22 |
| Ø1:Ø4:5Ø | TEMP | 27.9OFF | DO | 1.91 | NSTEP | 23 |
| Ø1:Ø4:55 | TEMP | 27.9OFF | DO | 1.93 | NSTEP | 22 |
| Ø1:Ø4:59 | TEMP | 27.9OFF | DO | 1.9Ø | NSTEP | 24 |
| Ø1:Ø5:Ø3 | TEMP | 28.2OFF | DO | 1.94 | NSTEP | 22 |
| Ø1:Ø5:Ø7 | TEMP | 28.1OFF | DO | 1.9Ø | NSTEP | 24 |
| Ø1:Ø5:12 | TEMP | 27.8OFF | DO | 1.94 | NSTEP | 22 |
| Ø1:Ø5:16 | TEMP | 28.2OFF | DO | 1.8Ø | NSTEP | 24 |
| Ø1:Ø5:25 | TEMP | 28.1OFF | DO | 1.9Ø | NSTEP | 27 |
| Ø1:Ø5:33 | TEMP | 27.8OFF | DO | 1.92 | NSTEP | 3Ø |
| Ø1:Ø5:37 | TEMP | 28.1OFF | DO | 1.91 | NSTEP | 31 |
| Ø1:Ø5:41 | TEMP | 28.1OFF | DO | 1.91 | NSTEP | 31 |
| Ø1:Ø5:45 | TEMP | 28.1OFF | DO | 1.92 | NSTEP | 3Ø |
| Ø1:Ø5:5Ø | TEMP | 28.1OFF | DO | 1.9Ø | NSTEP | 31 |
| Ø1:Ø5:58 | TEMP | 28.1OFF | DO | 1.91 | NSTEP | 33 |
| Ø1:Ø6:Ø2 | TEMP | 28.1OFF | DO | 1.91 | NSTEP | 33 |
| Ø1:Ø6:Ø6 | TEMP | 28.1OFF | DO | 1.89 | NSTEP | 34 |
| Ø1:Ø6:11 | TEMP | 27.8OFF | DO | 1.9Ø | NSTEP | 34 |
| Ø1:Ø6:15 | TEMP | 28.1OFF | DO | 1.92 | NSTEP | 33 |

**Table 3 : Data logging and results of temperature and dissolved oxygen control**

# CHAPTER 6

## RESULTS AND DISCUSSIONS

In the present study, temperature of the bioreactor was controlled using an ON/OFF control strategy and a comparative study was made for the control of dissolved oxygen concentration using P, P-I, P-I-D control schemes.

The temperature of the bioreactor was satisfactorily controlled using an ON/OFF control strategy as shown in Figure 7 to Figure 15.

A polarographic type of dissolved oxygen probe was used for the on-line monitoring of dissolved oxygen concentration in the bioreactor. The process transfer function was obtained experimentally using the Process Reaction Curve Method [30] and is found to be

$$K_p \quad \frac{e^{-t_d}}{\tau_p s + 1} \tag{22}$$

where

$$K_p = 0.002 \ \frac{\text{mg/l}}{\text{number of steps}} \tag{23}$$

$$t_d = 5 \text{ s.} \tag{24}$$

$$\tau_p = 47 \text{ s} \tag{25}$$

Cohen-Coon settings [28] were used to obtain the controller constants. On-line controller tuning was further done to obtain a satisfactory performance of the

control loop. The controller constants for the three control schemes are as given below:

Proportional control

$$K_c = 50 \frac{\text{number of steps}}{\text{mg/l}} \quad (26)$$

Proportional-Integral-Control

$$K_c = 40 \frac{\text{number of steps}}{\text{mg/l}} \quad (27)$$

$$\tau_I = 14 \text{ s.} \quad (28)$$

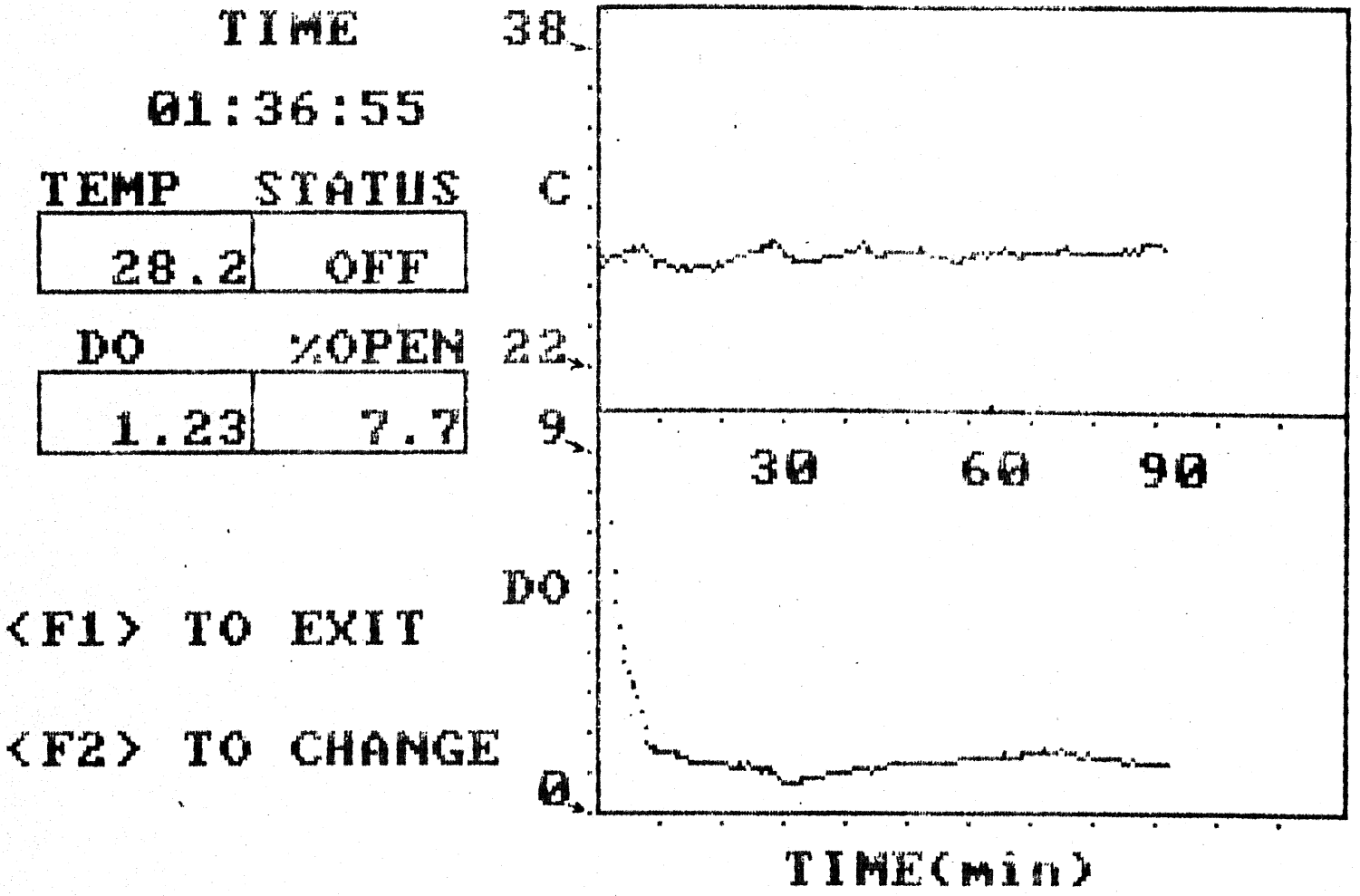
Proportional-Integral-Derivative Control

$$K_c = 65 \frac{\text{number of steps}}{\text{mg/l}} \quad (29)$$

$$\tau_I = 14.25 \text{ s} \quad (30)$$

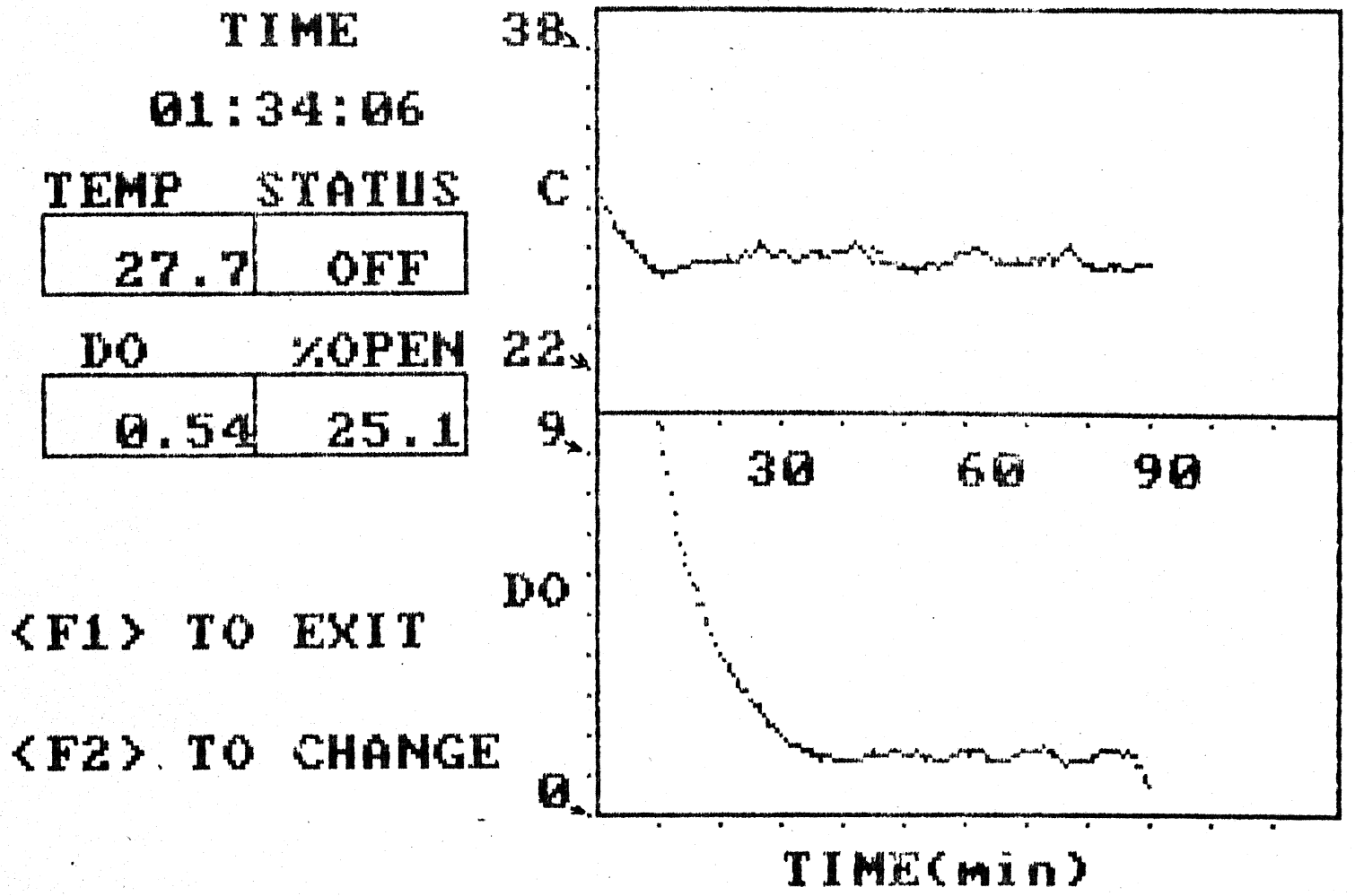
$$\tau_D = 1.78 \text{ s.} \quad (31)$$

Various experimental runs are shown in Figure 7 to Figure 15. Three sets of experimental runs were taken at setpoints - 1.5 mg/l, 2.0 mg/l, 2.5 mg/l of dissolved oxygen concentration. It was observed that the performance of the P, P-I, P-I-D control schemes was independent of the setpoint for the dissolved oxygen. Figures (7,10,13) show that the proportional control gives rise to an offset. Figures (8, 11, 14) show that the Proportional-Integral (P-I) Control removes the offset but gives rise to some oscillatory response. Proportional-Integral-Derivative (P-I-D) control was found to



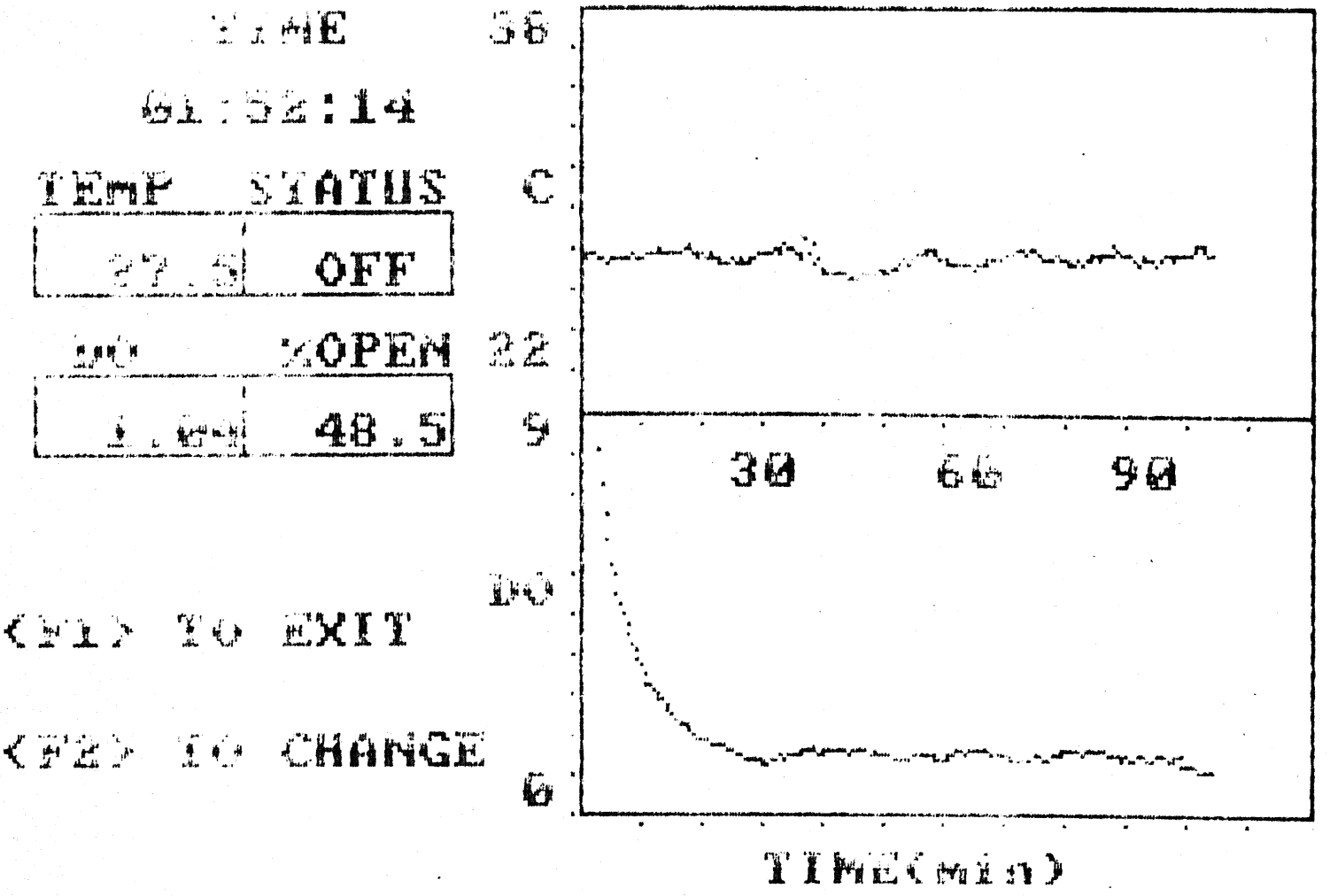

| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 1.5 | 0.1 |
| Temperature (°C) | 28 | 0.6 |

Fig. 7 Proportional control.



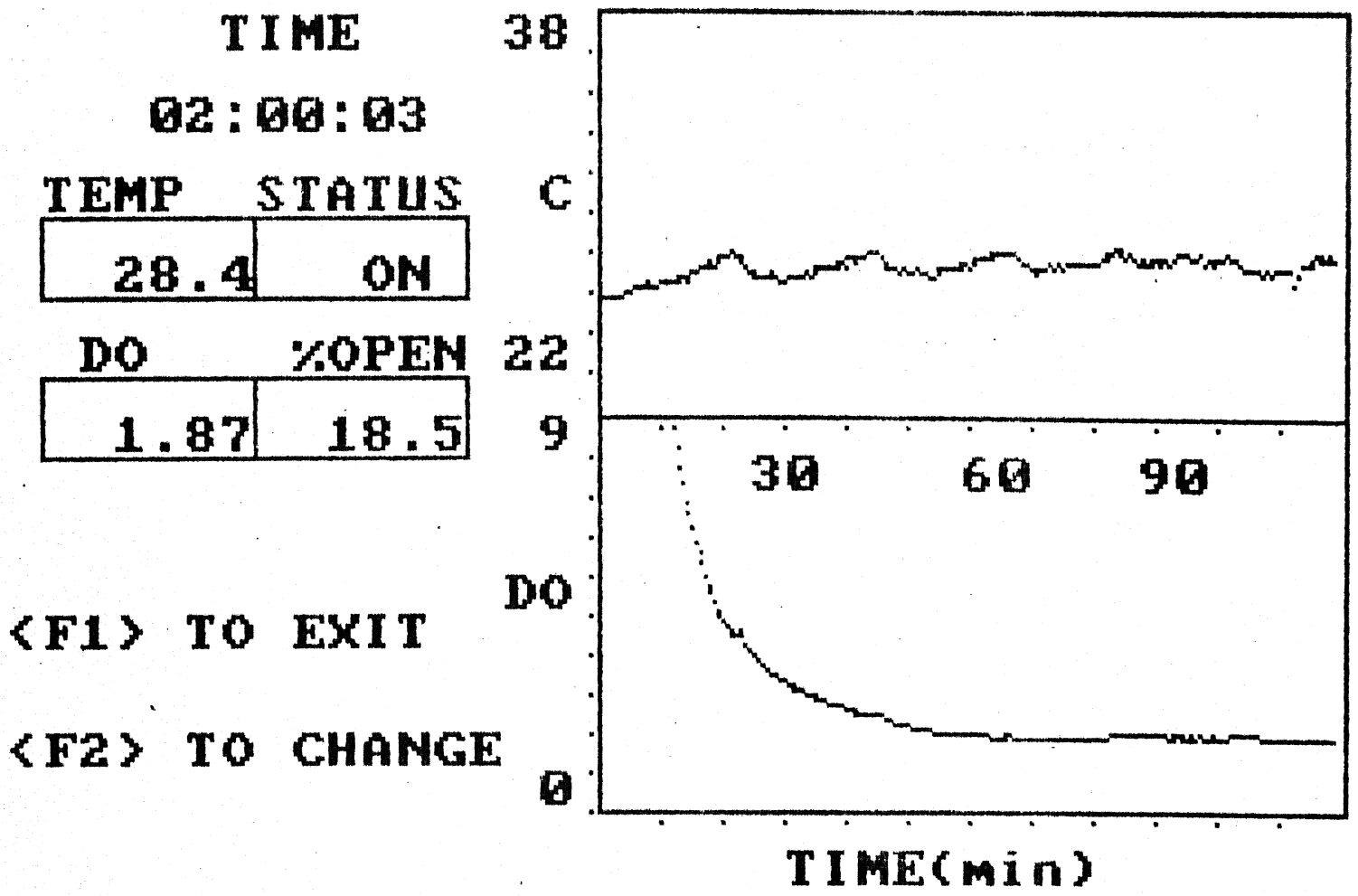

| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 1.5 | 0.1 |
| Temperature (°C) | 28 | 0.6 |

Fig. 8 Proportional - Integral (PI) control.

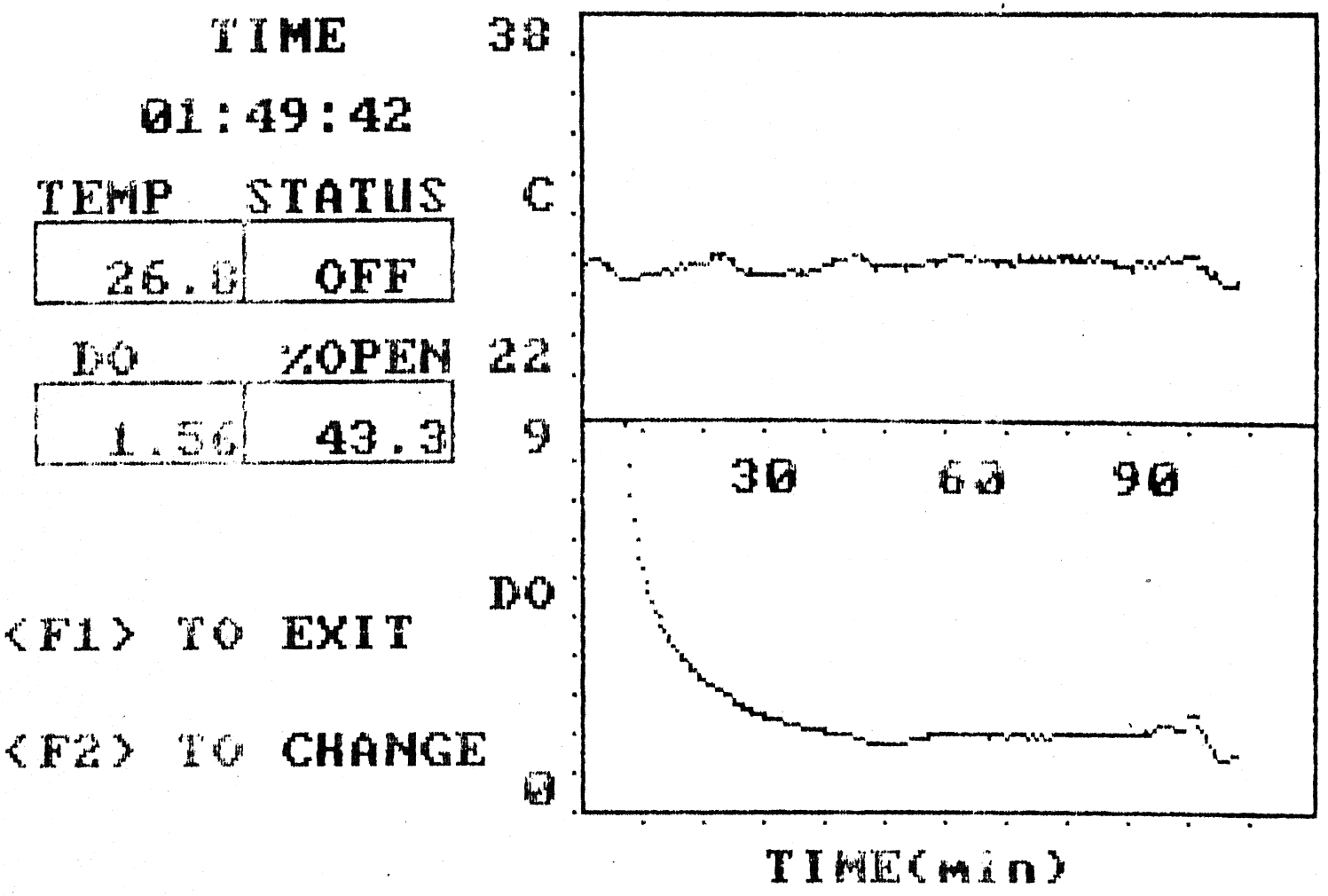



| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 1.5 | 0.1 |
| Temperature (°C) | 28 | 0.6 |

Fig. 9 Proportional-Integral-Derivative (PID) control.

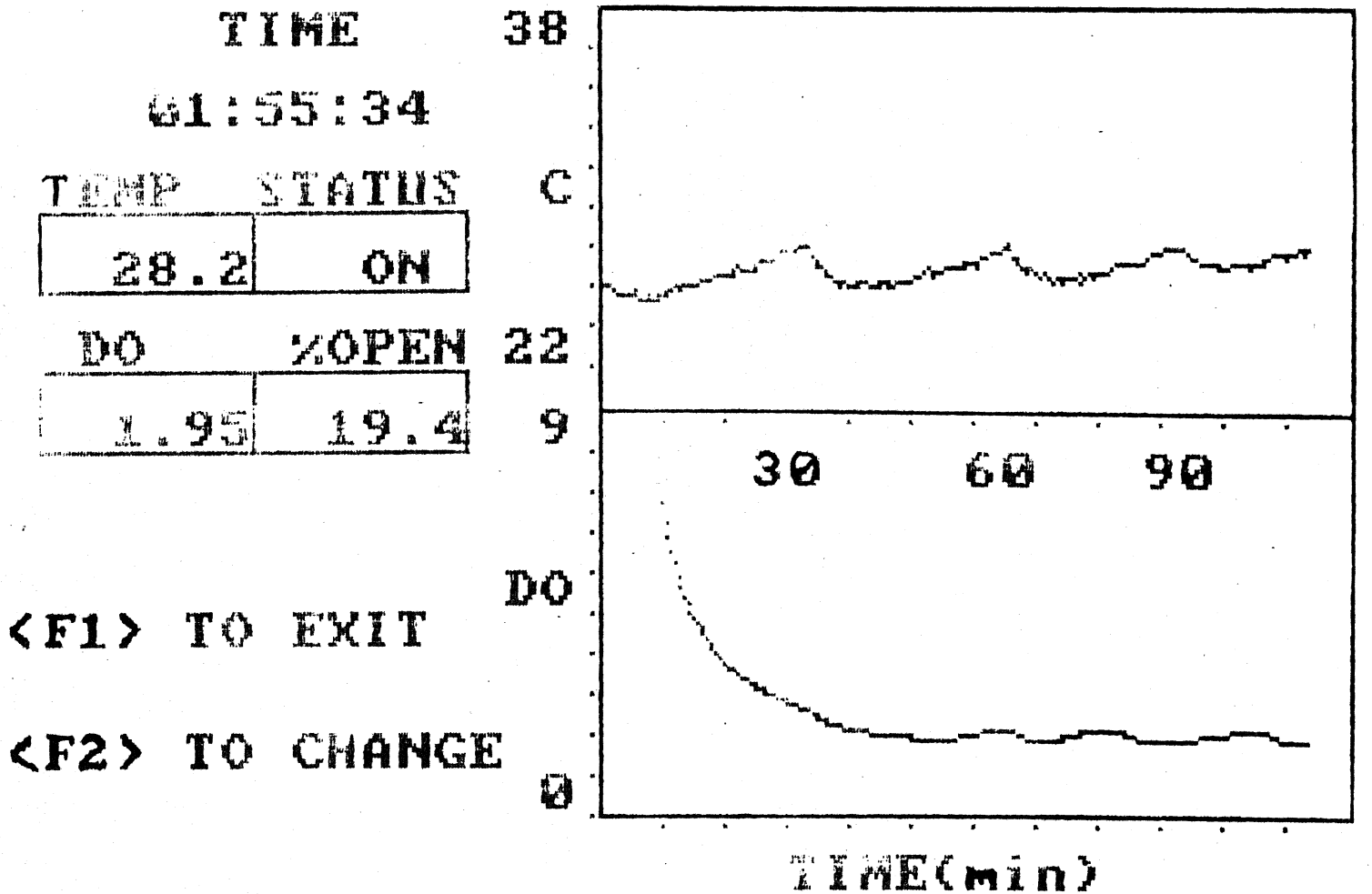



| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 2.0 | 0.1 |
| Temperature (°C) | 28 | 0.6 |



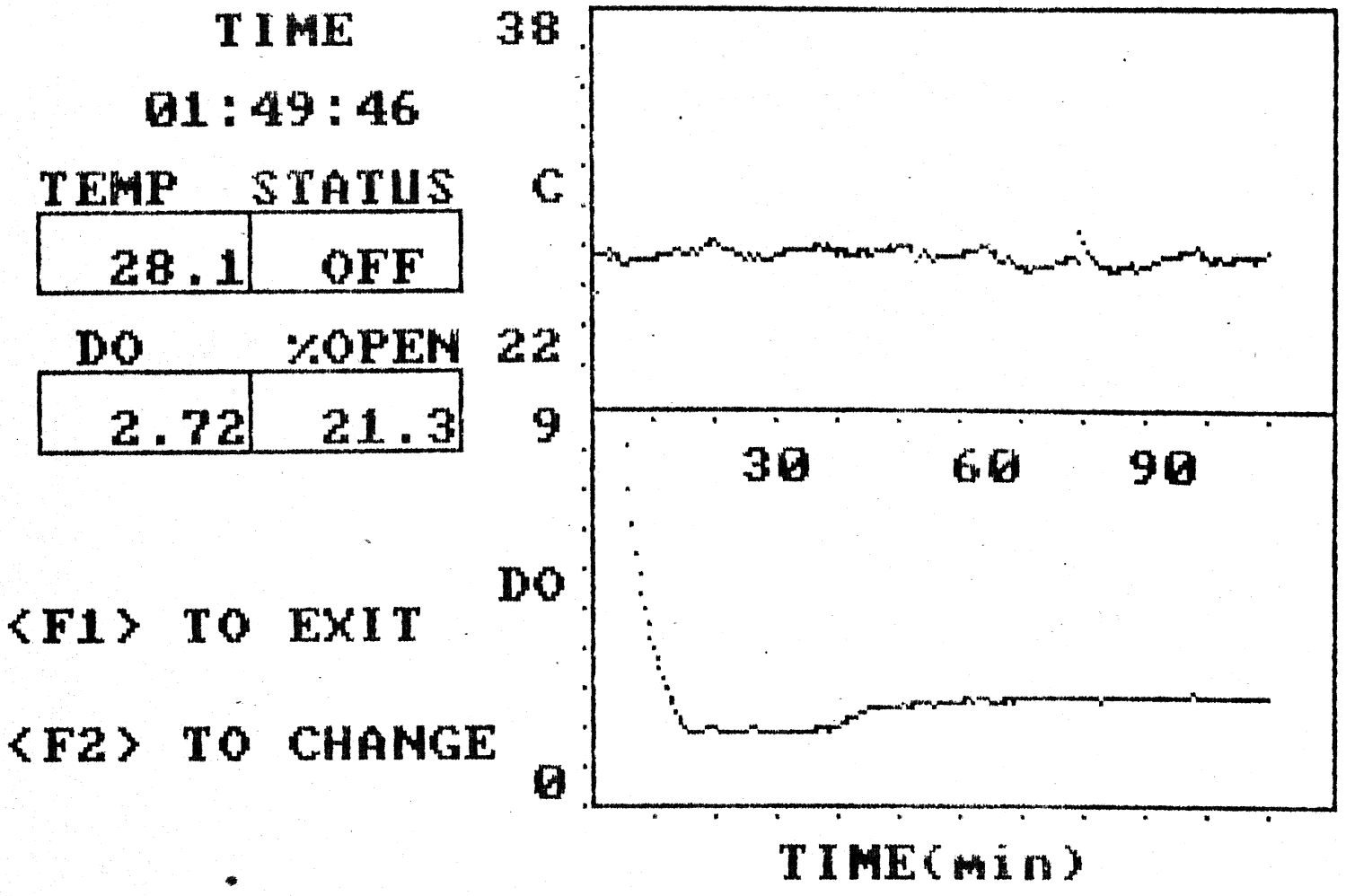

| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 2.0 | 0.1 |
| Temperature (°C) | 28 | 0.6 |

Fig. 11 Proportional-Integral control.

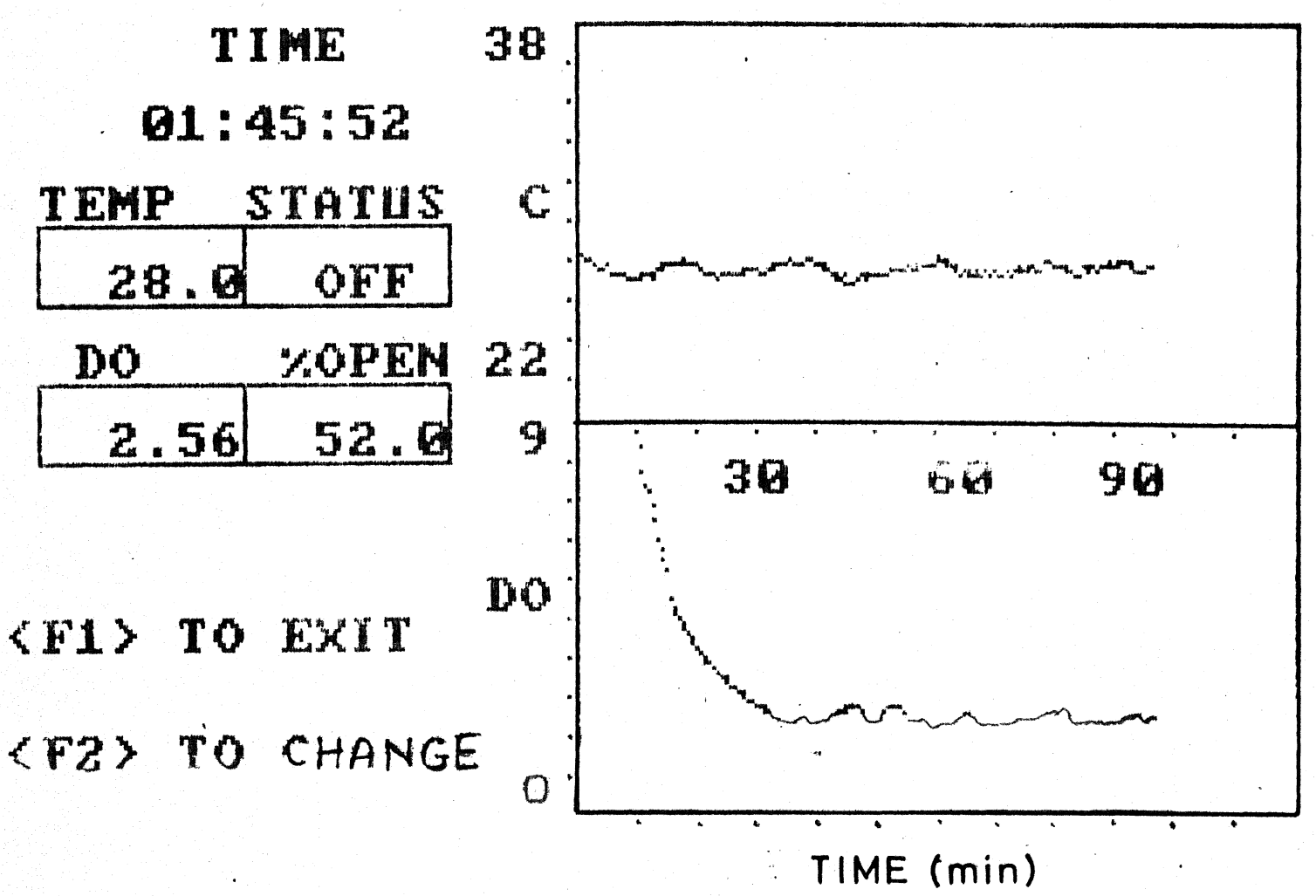



| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 2.0 | 0.1 |
| Temperature (°C) | 28 | 0.6 |

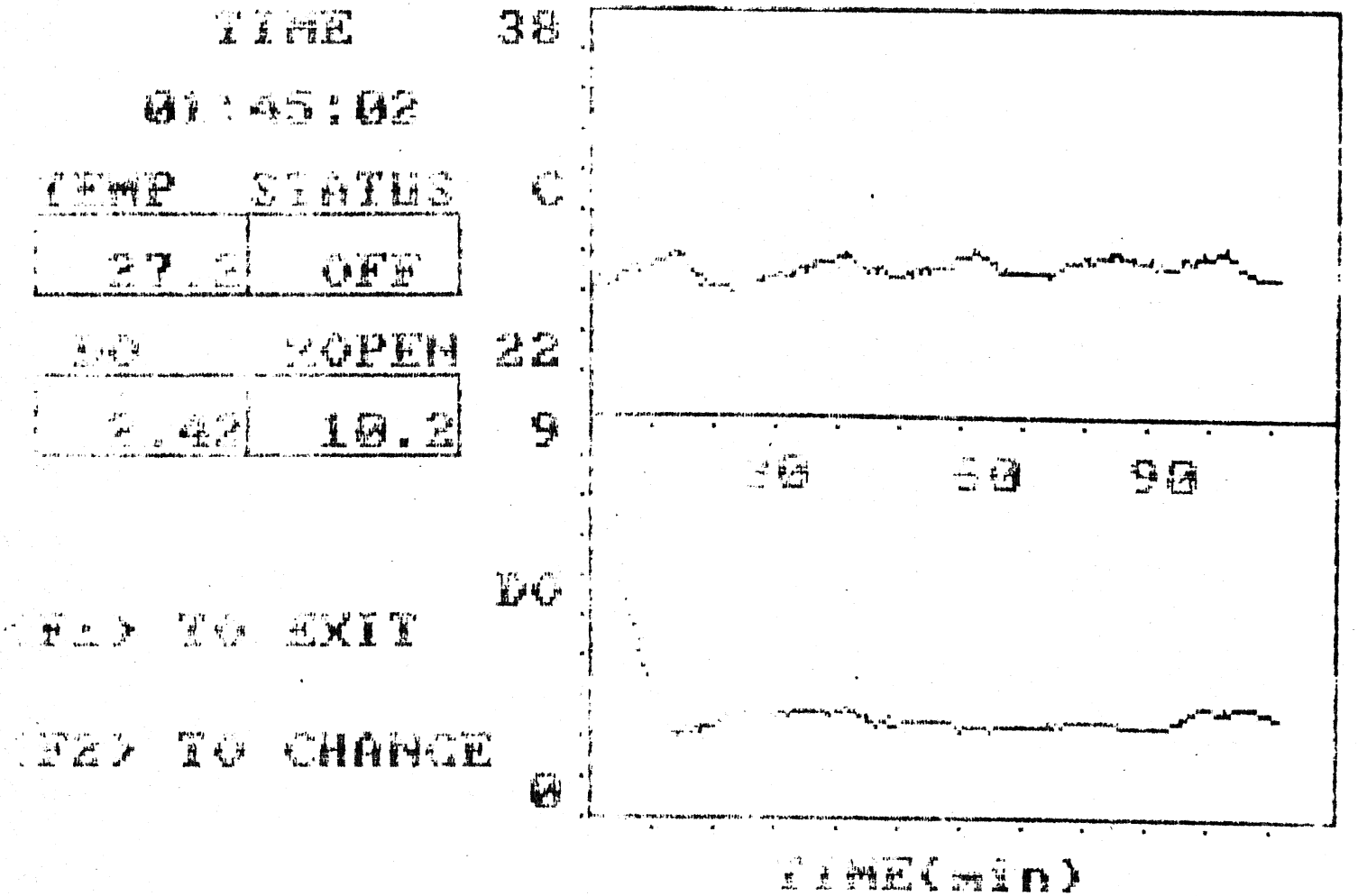
Fig. 12 Proportional - Integral - Derivative Control.




| | Setpoint | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 2.5 | 0.1 |
| Temperature (°C) | 28 | 0.6 |




| | Setpoint | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 2.5 | 0.1 |
| Temperature (°C) | 28 | 0.6 |




| | Setpoints | Deadband |
|---|---|---|
| Dissolved Oxygen (mg/l) | 2.5 | 0.1 |
| Temperature (°C) | 28 | 0.6 |

Fig. 15 Proportional - Integral - Derivative (PID) Control.

overcome both these defects as shown by (Figures 9, 12, 15). Higher controller gains as compared to P and P-I were possible (as shown by eqns. 26, 27, 29) which improved the speed of response of the system. For a case where the previous controller output was 50 steps of the step motor and the error at the nth, (n-1)th and (n-2)th instants were 0.5, 0.55 and 0.65 respectively the controller outputs for P, P-I, P-I-D controllers are -3, 4 and 15 steps respectively. Thus, the P-I-D algorithm has the quickest response since it will reach the setpoint at a faster rate as compared to the other two algorithms.

The behaviour of the probe was found to be erratic and there was a drop in the dissolved oxygen as indicated by the probe after 90 mins. The probe had to be washed with $NH_4OH$ and regenerated with saturated KCl solution before it could be reused.

This could be explained by the fact that the dissolved oxygen probe and membrances used for the present study were around 12 years old and hence the permeability and other properties of the polymeric membranes could have changed with time leading to an erratic behaviour.

The probe was also found to be unstable at higher levels of dissolved oxygen. Oneof the reasons for this could be that at the higher levels of dissolved oxygen, the electrolyte (saturated KCl solution) gets depleted at a rapid rate. The

probe had to be regenerated for reuse. This problem can be eliminated by using a set of probes which can be used alternatively.

# CHAPTER 7

## CONCLUSIONS AND RECOMMENDATIONS

1. The temperature of the bioreactor was satisfactorily controlled using an ON/OFF control strategy.

2. A comparative study of Proportional, Proportional-Integral and Proportional-Integral-Derivative Control schemes was carried out for the control of dissolved oxygen concentration. The Proportional-Integral-Derivative (PID) strategy had the quickest response and eliminated offset and hence is the most suitable strategy amongst P, P-I and P-I-D for the control of the dissolved oxygen concentration.

3. The dissolved oxygen probe exhibited an erratic behaviour and there was a drop in the value of the dissolved oxygen as indicated by the probe after a period of 90 mins. due to theprobe and polymeric membranes being more than 12 years old. Electrical interference was observed between the dissolved oxygen probe and the other two sensors, viz. the thermocouple and the pH probe. As soon as the dissolved oxygen probe was inserted into the broth both the temperature and the pH signals went haywire. This was due to the electrical inter-connection of the probes through the broth. The

thermocouple was electrically isolated by placing it in a thin, closed glass tube. No such isolation was possible for the pH probe because the liquid must touch its glass bulb for pH measurement. Thus, it was not possible to run the dissolved oxygen probe and the pH probe simultaneously.

4. The present study can be used to develop a single board computer based dedicated control system with a view to further reduce the cost to approximately Rs.8000. As the software has already been tested on the Usha Eagle PC, it can be transferred to a ROM or to an EPROM. The program in the ROM may contain the default values for temperature, pH and dissolved oxygen. Set point changes can be achieved by the software residing in the RAM. Alternatively, the software can be written as to require the user to input the setpoints before each run. Also a warning can be flashed if any of the variables exceed the desired range. A four or six digit display can be used to display the values of the controlled variables and the valve status. Thus, this system will not have an on-line display to monitor the progress of the fermentation. The values of the controlled variables can be printed on an on-line printer. If additional variables are to controlled in future, the EPROM has to be re-programmed.

5. The software can be modified to control parameters in the chemical process industry.

RECOMMENDATIONS:

1. Multivariable control strategies which consider interaction amongst temperature, pH and dissolved oxygen can be studied. This will fulfil the need for a comprehensive control scheme for bioreactors. The saturation level of dissolved oxygen decreases with temperature but the volumetric mass transfer coefficient for oxygen transfer increases with temperature and hence these effects tend to cancel each other. Experimental studies need to be carried out to determine the interaction between temperature, pH and dissolved oxygen. Decoupling of these variables can be done to eliminate the effect of interaction amongst them.

2. The interaction between the pH and the dissolved oxygen probe can be eliminated by using an optically isolated interface instead of DT-707T. The output of the dissolved oxygen probe can be fed to a Parallel Interface Adapter (PIA) chip through an ADC. The outputs from the PIA chip are connected to the computer through optical isolators. An alternative, would be to use a galvanic dissolved oxygen probe which does not require a polarizing voltage.

3. A dedicated control system should be set up to reduce the cost.

4. More parameters (eg., exit oxygen concentration, exit carbon dioxide concentration, etc.) can be monitored and thus indirect monitoring and control of the biomass can be made.

5. This system can be used to control and optimise the fermentation of other substrates.

6. Software can be further developed to control intricate chemical processes.

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# APPENDIX 1

## SPECIFICATIONS OF USHA EAGLE PC

8088 Intel microprocessor at 4.77 MHz

640 KB RAM

Colour graphics card with monochrome monitor

2x360 KB floppy drives

EPSON FX-80 dot matrix printer

Data Translation DT-2805 Low Level Analog and Digital Interface card with DT-707T screw termination panel.

# APPENDIX 2

## SPECIFICATIONS OF DT-2805 CARD

Analog Inputs:

| | | |
|---|---|---|
| No. of channels | : | 8 Differential |
| Resolution | : | 12 Bits |
| Gain | : | 1,10,100,500 Software selectable |
| Input voltage | : | ± 10 volts (maximum) |

Analog Outputs:

| | | |
|---|---|---|
| No. of channels | : | 2 |
| Resolution | : | 12 bits |
| Output voltage | : | ± 10 volts (maximum) |
| A/D conversion time | : | 25 microseconds |
| A/D throughput to system memory | : | 6000 sps. |
| Digital Input/Output | : | 16 channels |

The DT-707T is its companion screw termination panel with room temperature sensing hardware. It is an accessory product which permits all user connections (A/D, D/A, Digital I/O, External Trigger and External Clock) to the DT-2805 board.

# APPENDIX 3

## CALIBRATION OF DISSOLVED OXYGEN PROBE

The Winklers Titration Method [29] was used to calibrate the dissolved oxygen probe. The dissolved oxygen concentration of the sample was obtained chemically and the corresponding millivolt signal from the dissolved oxygen probe was noted.

The reagents required for the determination of dissolved oxygen concentration are:

(1) Manganese sulfate solution

436 g $MnSO_4.4H_2O$ or 400g $MnSO4.2H_2O$

is dissolved in 800 ml distilled water, filtered if required and diluted to one litre.

(2) Alkali-Iodide-Azide Reagent

500 gms NaOH and 150 gms sodium iodide are dissolved in distilled water. 10 gms sodium azide dissolved in 40 ml distilled water is added to this solution and is diluted to one litre.

(3) Conc. $H_2SO_4$.

(4) Starch Indicator

5 gms. is dissolved in 800 ml boiling water; cooled and diluted to one litre.

(5) Standard sodium thiosulfate solution (0.025N)

The procedure for determining the dissolved oxygen concentration is as follows:

(1) To the BOD bottle with sample, add 2 ml $MnSO_4$ followed by 2 ml alkali-iodide-azide reagent by inserting the pipette well below the surface of the liquid. Close the bottle tightly and mix the contents thoroughly by inverting the bottle atleast six times.

(2) After letting it settle for 2 minutes, remove the stopper and add 2 ml conc. $H_2SO_4$. Stopper the bottle and mix by gentle inversion, till the dissolution of the precipitate is complete and there is an uniform distribution of the liberated iodine.

(3) Take 203 ml from the bottle and titrate with standard thiosulfate to a pale straw colour. Add 5 ml starch solution, continue titration till the blue colour, continue titration till the blue colour disappears.

(4) The volume of titrant (ml) gives the dissolved oxygen concentration in mg/l.

The calibration plot of dissolved oxygen (mg/l) versus millivolts is given in Figure 16 and is found to be a straight line.

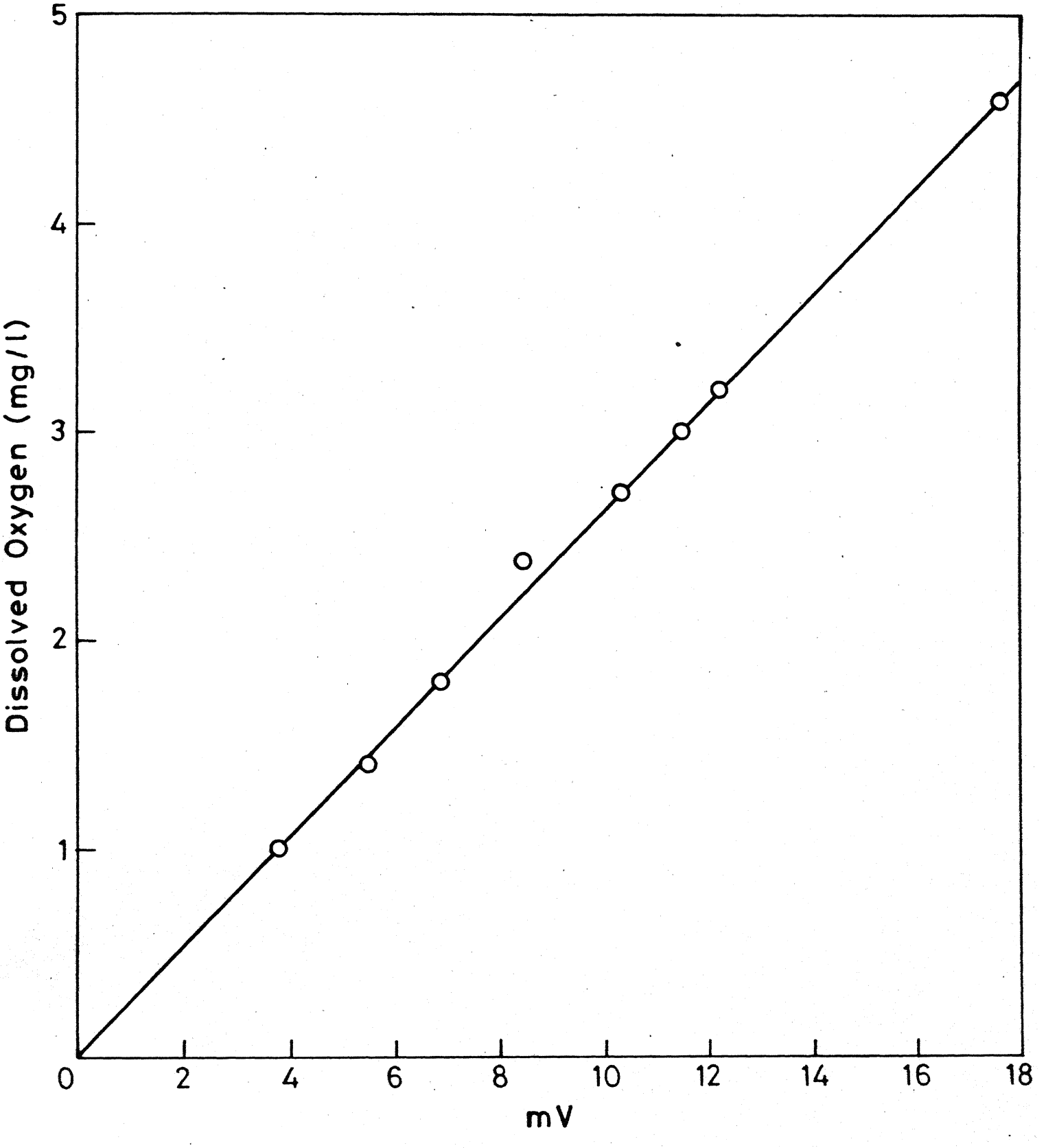


Fig. 16 Calibration plot for D.O. probe .

# APPENDIX 4

## PROCESS REACTION CURVE METHOD [30]

The Process Reaction Curve Method is used to experimentally determine the process transfer function. A step change of magnitude A is introduced in the variable C which actuates the final control element. The value of the output is recorded with respect to time. The process gain, process time constant and dead time are then obtained as follows:

$$\text{Process gain} = \frac{\text{Steady state change in output}}{\text{Change in input}}$$

Process Time Constant: Time required to reach 63.2% of final value in response to a fixed (i.e. step) change in input.

Dead Time: Time elapsed until the system responded.

For the present system, a step input of 900 steps was given in the step motor driven needle valve thereby increasing the airflow rate. The value of dissolved oxygen was then recorded with respect to time. The initial steady state in dissolved oxygen was 2.0 mg/l and the final steady state was 3.8 mg/l.

Thus the process gain is given by,

$$K_p = \frac{3.8 - 2.0}{900} = 0.002.$$

The process time constant was calculated from the response curve (Figure 17) and found to be 47 secs. The dead time was observed to be 4 secs.

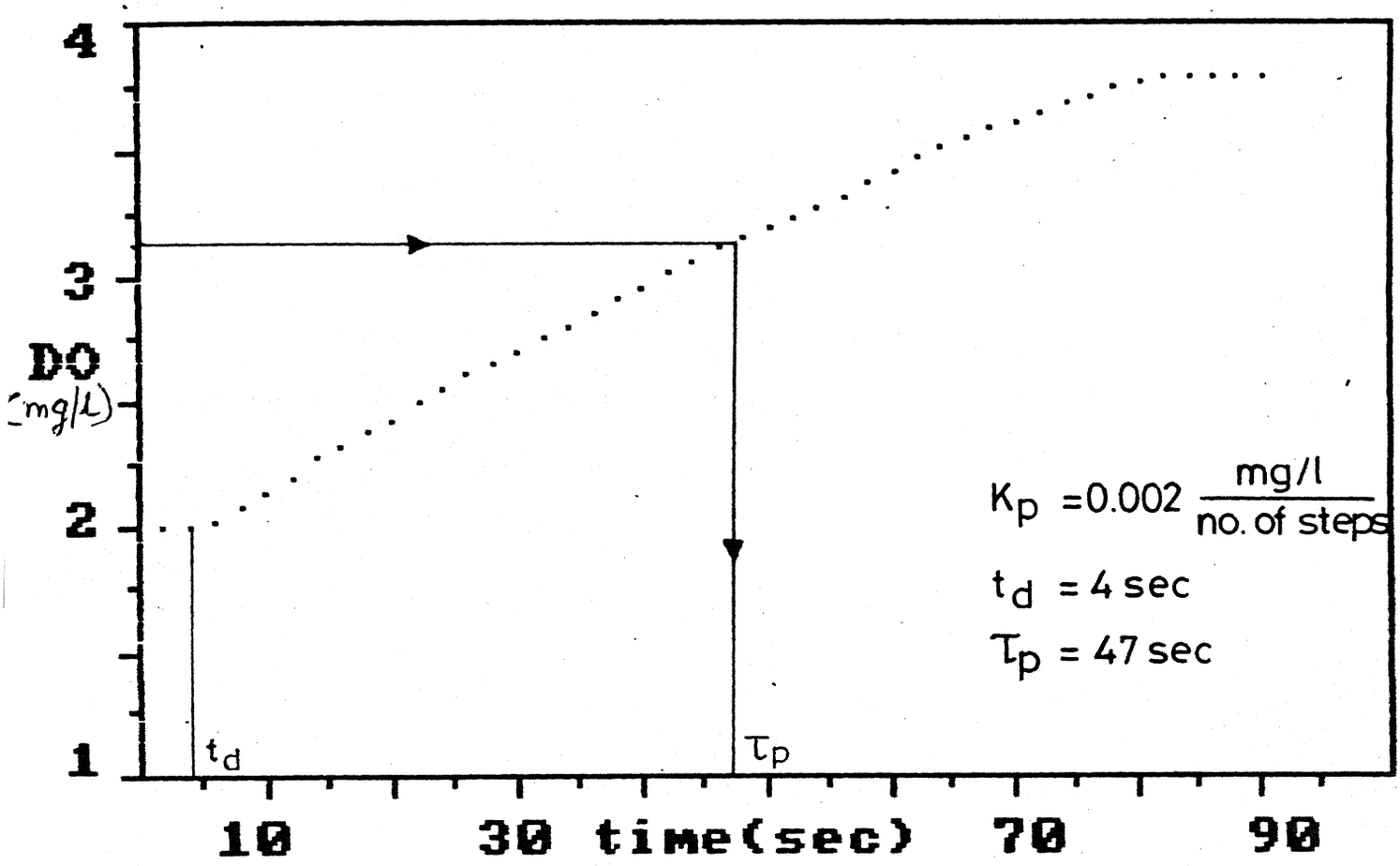


Fig. 17 Process reaction curve to obtain the process transfer function.

Listing of the Program

```
10 SCREEN 1: CLS
20 PRINT : INPUT "INSERT pctherm dISK,READY PRINTER & HIT RETURN";DUM$
30 DEF SEG=7552
40 BLOAD "PCTHERM",0
50 OPEN"o",#1,"b:OUTPUT"
60 OPEN "o",#2,"b:GROUTPUT"
70 GOSUB 940
80 CLS:PRINT
90 '*****************************************
100 ' INPUT SETPOINTS FOR TEMP,DISS. OXYGEN *
110 '*****************************************
120 CALL XMT(TYPE%,CHTEMP%,TEMP)
130 PRINT "BATH TEMPERATURE (C) :";
140 PRINT USING "###.#";TEMP
150 INPUT "SETPOINT (C)  :",SETPOINT
160 INPUT "VARIATION  :",DEL
170 DELTA=DEL/2
180 TMAX=SETPOINT+DELTA
190 TMIN=SETPOINT-DELTA
200 PRINT
210 INPUT "SETPOINT IN DO(mg/l):",DOSET
220 INPUT "VARIATION           :",DELTADO
230 DELTAD=DELTADO/2
240 DOMAX=DOSET+DELTAD
250 DOMIN=DOSET-DELTAD
260 PRINT
270 CLS
280 PRINT "CHOOSE A CONTROLLER ALGORITHM FROM THE FOLLOWING "
290 PRINT "1 FOR PROPORTIONAL"
300 PRINT "2 FOR P-I"
310 PRINT "3 FOR P-I-D"
320 INPUT "TYPE=";TYPE
330 IF TYPE=1 THEN GOSUB 2940
340 IF TYPE=2 THEN GOSUB 3000
350 IF TYPE=3 THEN GOSUB 3070
360 INPUT "Hit <ENTER> TO START",DUM$
370 PRINT#1,"setpoints";CHR$(13);SPC(10);"TEMPERATURE(C)";SETPOINT;SPC(5);
    "DEADBAND";DEL
380 PRINT#1,SPC(10);"DO";DOSET;"DEADBAND";DELTADO
390 LPRINT,"setpoints";CHR$(13);SPC(10);"TEMPERATURE(C)";SETPOINT;SPC(5);
    "DEADBAND";DEL
400 LPRINT,SPC(10);"DO";DOSET;"DEADBAND";DELTADO
410 CLS
420 KEY OFF
430 GOSUB 1310
440 KEY(1) ON : ON KEY(1) GOSUB 1740
450 KEY(2) ON : ON KEY(2) GOSUB 1800
```

```
460 '*************************************
470 '     CONTROL                         *
480 '*************************************
490 Z=TIMER:N=0:SUMDO=0:SUMT=0
500 WHILE Z+30>TIMER
510 LOCATE 4,6
520 T$=TIME$ : PRINT T$
530 GOSUB 2070 'temp. control
540 'GOSUB 2130 'ph control
550 IF DIGITAL.VALUE%=DV%+PV% THEN 590
560 DIGITAL.VALUE%=DV%+PV%
570 CALL XODV(DVPORT%,DVMASK%,DIGITAL.VALUE%)
580 GOSUB 1840
590  FOR I=1 TO 200
600  CALL XMV(CHDO%,DOVOLTS)
610  DOMV=DOMV+DOVOLTS*1000
620  NEXT
630   DOMV=DOMV/200
640   DO=.26315*DOMV
650   LOCATE 12,4
660   PRINT USING "##.##";DO
670 IF DO<DOMIN THEN IND=IND+1
680 IF IND>0 THEN GOSUB 2350
690 PCENT=NSTEP/18
700 LOCATE 12,10
710 PRINT USING "###.#";PCENT
720 SOUND 32767,18
730 SUMDO=SUMDO+DO
740 SUMT=SUMT+TEMP
750 N=N+1
760 WEND
770 AVGDO=SUMDO/N
780 AVGT=SUMT/N
790 J=J+1
800 K=J*30
810 VIEW (142,7)-(311,88)
820 WINDOW (0,20)-(7200,40)
830 PSET(K,AVGT)
840 VIEW (142,88)-(311,168)
850 WINDOW (0,0)-(7200,10)
860 PSET (K,AVGDO)
870  PRINT #2,USING "##.#";AVGDO;
880 PRINT #2, SPC(2);
890 PRINT #2, USING "##.#";AVGT;
900 PRINT #2, SPC(2);
910 PRINT #2,K
920 GOTO 490
930 '*************************************
940 '          INITIALISATION             *
950 '*************************************
960 DEFINT V
970 DEFINT X: DEFINT I-N
980 XGV=165 'xgv(dac.select%,volts!)-to generate volts
990 XMV=138 'xmv(ch%,volts)-to measure volts
1000 XEFO=60 'xefo(port%)-enable for output
```

```
1010 XODV=66 'xodv(port%,mask%,digital.value%)-output digital value
1020 XRD=111 'xrd(dt%)-reset board
1030 XMCJ=144 'measure cold jn.
1040 XMT=141 'xmt(type%,ch%,temp)-to measure thermocouple
1050 DT%=0
1060 CALL XRD(DT%)
1070 DVPORT%=0:STPORT%=1
1080 DAC.SELECT%=1
1090 VOLTS!=.835 'TO GENERATE 0.8 VOLTS
1100 CALL XGV(DAC.SELECT%,VOLTS!)
1110 CALL XEFO(DVPORT%)
1120 CALL XEFO(STPORT%)
1130 DVMASK%=&H3:STMASK%=&HF
1140 CHDO%=5:CHTEMP%=1:CHPH%=4
1150 TYPE%=ASC("T")
1160 DIGITAL.VALUE%=&H0
1170 CALL XODV(DVPORT%,DVMASK%,DIGITAL.VALUE%)
1180 COLDON%=&H1:PHON%=&H2
1190 BOTHOFF%=&H0
1200 NSTEP=0:IND=0
1210 J=0
1220 DIM A(4)
1230 VOLD=0
1240  ERRP=0 : ERRP1=0
1250  KPRC=.002:TDEAD=4:TPRC=47:TSAMP=4
1260 '*******************************
1270 'KPRC:PROCESS GAIN
1280 'TDEAD : DEAD TIME
1290 'TPRC:PROCESS TIME CONSTANT
1300 'TSAMP : SAMPLING RATE
1310 '*******************************
1320 RETURN
1330 '************************************
1340 '          PLOTTING SUBROUTINE      *
1350 ' ***********************************
1360 LINE (16,56)-(112,72),,B
1370 LINE (16,88)-(112,104),,B
1380 LINE (142,15)-(311,176),,B
1390 LINE (64,56)-(64,72)
1400 LINE (64,88)-(64,104)
1410 LOCATE 3,8,0 : PRINT "TIME"
1420 LOCATE 7,3,0 : PRINT "TEMP"
1430 LOCATE 7,9,0 : PRINT "STATUS"
1440 LOCATE 11,3,0 : PRINT " DO"
1450 LOCATE 11,10,0 : PRINT "%OPEN"
1460 LOCATE 24,24,0: PRINT "TIME(min)"
1470 LOCATE 16,16,0 : PRINT "DO"
1480 LOCATE 6,17,0 : PRINT "C"
1490 LOCATE 2,16,0 : PRINT "38"
1500 LOCATE 10,16,0: PRINT "22"
1510 LOCATE 12,17,0 : PRINT "9"
1520 LOCATE 21,17,0 : PRINT "0"
1530 LOCATE 13,23,0 : PRINT "30"
1540 LOCATE 13,29,0 : PRINT "60"
1550 LOCATE 13,34,0 : PRINT "90"
```

```
1560 LOCATE 17,2
1570 PRINT "<F1> TO EXIT"
1580 LOCATE 20,2
1590 PRINT "<F2> TO CHANGE"
1600 FOR I=15 TO 79 STEP 8
1610 LINE (140,I)-(142,I),,,&HA0A0
1620 NEXT
1630 LINE (142,88)-(311,88)
1640 FOR I= 96 TO 168 STEP 8
1650 LINE (140,I)-(142,I),,,&HA0A0
1660 NEXT
1670 FOR I=156 TO 297 STEP 14
1680 LINE (I,88)-(I,90),,,&HA0A0
1690 NEXT
1700 FOR I=156 TO 297 STEP 14
1710 LINE (I,168)-(I,172),,,&HA0A0
1720 NEXT
1730 RETURN
1740 KEY ON
1750 CLOSE#1
1760 CLOSE#2
1770 CALL XRD(DT%)
1780  SCREEN 0,0,0 :CLS
1790 END
1800  DIGITAL.VALUE%=&H0
1810 SCREEN 0,0,0 :CLS
1820 SCREEN 1 : CLS
1830 GOTO 110
1840 ' ***********************************
1850 '                    LINE PRINTING  *
1860 ' ***********************************
1870 LPRINT T$;SPC(2);
1880 LPRINT "TEMP";SPC(2);
1890 LPRINT USING "###.#";TEMP;
1900 IF DV%=0 THEN LPRINT "OFF";SPC(2); ELSE LPRINT "ON";SPC(2);
1910 LPRINT "DO";SPC(2);
1920 LPRINT USING "##.##";DO,
1930 LPRINT SPC(2);
1940 LPRINT "NSTEP";SPC(4);
1950 LPRINT USING "####";NSTEP
1960 T$=TIME$
1970  PRINT#1, T$;SPC(2);
1980  PRINT#1, "TEMP";SPC(2);
1990  PRINT#1, USING "###.#";TEMP;
2000 IF DV%=0 THEN  PRINT#1, "OFF";SPC(2); ELSE  PRINT#1," ON";SPC(2);
2010  PRINT#1, "DO";SPC(2);
2020  PRINT#1, USING "##.##";DO;
2030  PRINT#1,SPC(2);
2040  PRINT#1, "NSTEP";SPC(4);
2050  PRINT#1, USING "####";NSTEP
2060 RETURN
```

```
2070 '*******************************
2080 ' temperature control       *
2090 '*******************************
2100  CALL XMT(TYPE%,CHTEMP%,TEMP)
2110  LOCATE 8,4
2120 PRINT USING "###.#";TEMP
2130 IF TEMP>TMAX THEN DV%=COLDON%
2140 IF TEMP<TMIN THEN DV%=BOTHOFF%
2150 LOCATE 8,11
2160 IF DV%=COLDON% THEN PRINT " ON" ELSE PRINT "OFF"
2170 RETURN
2180 '*****************
2190 'pH control      *
2200 '*****************
2210 PHMV=0
2220 FOR I=1 TO 10
2230 CALL XMV(CHPH%,PHVOLTS)
2240 PHMV=PHMV+PHVOLTS*1000
2250 NEXT
2260 PHMV=PHMV/10
2270 PH=-1.103346*PHMV+6.585139
2280 IF PH<PHMIN THEN PV%=PHON%
2290 IF PH>PHMAX THEN PV%=BOTHOFF%
2300 LOCATE 13,4
2310 PRINT USING "##.##";PH
2320 LOCATE 13,11
 30 IF PV%=PHON% THEN PRINT " ON" ELSE PRINT "OFF"
 40 RETURN
 50 '***********************
 60 '   do control        *
 70 '***********************
 80 DDIFF=DOSET-DO
 90  V=VOLD+(CC)*((DDIFF-ERRP)+(X*(TSAMP/TI)*DDIFF)+(Y*(TD/TSAMP)*(DDIFF-2*ERRP+ERRI
00  ERRP1=ERRP
10  ERRP=DDIFF
20  IF DO>DOMAX THEN GOSUB 2480
30  IF DO<DOMIN THEN GOSUB 2480
40  PCENT=NSTEP/18
50  LOCATE 12,10
50  PRINT USING "###.#";PCENT
70  RETURN
80  DIFF=V-VOLD
90 IF DIFF>0 THEN GOSUB 2530
00  IF DIFF<0 THEN GOSUB 2710
 0 GOSUB 1840
 0 RETURN
 0 '*******************************
 0 ' forward direction(9-5-6-a) *
 0 '*******************************
 0  IF V>1800 THEN DIFF=1800-VOLD
 0  A%(0)=&H9
 0  A%(1)=&H5
 0  A%(2)=&H6
 0  A%(3)=&HA
 0  NDIFF=DIFF/4
```

```
2620  FOR I=1 TO NDIFF
2630     FOR X=0 TO 3
2640     CALL XODV(STPORT%,STMASK%,A%(X))
2650     NEXT
2660  NEXT
2670  V=DIFF+VOLD
2680  VOLD=V
2690 NSTEP=V
2700  RETURN
2710 '*********************************
2720 '   reverse direction(to shut) *
2730 '*********************************
2740  IF V<0 THEN DIFF=-VOLD
2750  A%(0)=&H6
2760  A%(1)=&H5
2770  A%(2)=&H9
2780  A%(3)=&HA
2790  NDIFF=-DIFF/4
2800  FOR I=1 TO NDIFF
2810     FOR X=0 TO 3
2820     CALL XODV(STPORT%,STMASK%,A%(X))
2830     NEXT
2840  NEXT
2850  V=VOLD+DIFF
2860  VOLD=V
2870 NSTEP=V
2880  RETURN
2890 '*********************************
2900 'CC : PROPORTIONAL GAIN
2910 'TI : INTEGRAL TIME CONSTANT
2920 'TD : DERIVATIVE TIME CONSTANT
2930 '*********************************
2940 '*****************************************
2950 'SETTINGS FOR PROPORTIONAL CONTROLLER *
2960 '*****************************************
2970 CC=50
2980 X=0 : Y=0
2990 RETURN
3000 '*********************************
3010 'SETTINGS FOR P-I CONTROLLER *
3020 '*********************************
3030 CC=40
3040 TI=14
3050 X=1 : Y=0
3060 RETURN
3070 '*********************************
3080 'SETTINGS FOR P-I-D CONTROLLER*
3090 '*********************************
3100 CC=65
3110 TI=14.25
3120 TD=1.7836
3130 X=1 : Y=1
3140 RETURN
3150 '*********************************
```